

Selection of Some Female Date Palm Trees from Dakhla Oasis New Valley Governorate

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ABSTRACT

Two seasons in a row (2023-2024) were consumed for this inquiry at El-Wahat El-Dakhla in the governorate of the New Valley. There were two sections of the current study. The first goal was to choose date palm genotype trees from the specified site that had high-quality fruit. Six semi-dry genotypes date palms were examined for their fruit's physical and chemical characteristics in comparison to the Sultany variety. Date palm genotype No. 3 (97.66 units), No. 1 (91.27 units), No. 4 (86.78 units), No. 6 (84.4 units), No. 2 (82.68 units), No. 5 (81.46 units), and Sultany cultivar (63.56 units) were the results of the numerical selection of the chosen palms. The second objective of this study is to use inter-simple sequence repeats (ISSR) as a DNA fingerprint to identify the genetic diversity among chosen genotypes.

Keywords: Date palm, Semi-dry genotypes, DNA, ISSR.

INTRODUCTION

One of the most significant and ancient tree fruits that humans have ever cultivated is the date palm (*Phoenix dactylifera* L.), which is also thought to be the oldest tree with the greatest genetic diversity (Popenoe, 1973). The New Valley and El-Dakhla Oasis have been reported to be favorable environment for growing and fruiting date palms, particularly soft and semi-dry fruit varieties. In the Arab Republic of Egypt, this crop consistently ranks higher than many other fruit crops (Hussein *et al.*, 1979).

Due to sexual reproduction, date palms had produced a large number of genotypes, some of which are very attractive for their fruit quality and yield. The selection or assessment of seedy date palms was the subject of several studies (Abdalla, 2002 and Mostafa, 1986).

The main objective of this study was to figure out which genotype trees were considered the most promising based on their productivity and fruit quality, which was determined by examining the fruit's yield as well as physical characteristics and chemical composition.

An ability to select promising plant material in order to speed up the selection process is essential for the genetic advancement of a crop species. For a selection technique, molecular markers relating to significant qualities may be employed. The markers can then be utilized to create genetic maps, which are valuable resources for in-depth genetic study and more precise marker-assisted breeding strategies utilizing selection (Soliman *et al.*, 2006). Hence, in order to access the DNA diversity among crop genotypes, using ISSR approach was utilized (Zehdi *et al.*,

2004). Thus, the second objective of this study is to use inter-simple sequence repeats (ISSR) as a DNA fingerprint to identify the genetic diversity among chosen genotypes.

MATERIALS AND METHODS

Two seasons (2023-2024) were utilized to conduct this study in the orchards of El-Wahat El-Baharia oasis in the governorate of New Valley. Six semi-dry date palm plants were selected while; The Sultany cultivar tree was chosen as a common comparison across numerous excellent genotypes palms.

The chosen palms had good production and excellent fruit qualities; they were around 25 years old and had been planted in sandy soil. Each tree has a consistent growth pattern and is in good physical shape. They experienced the same cultural customs. To prevent the impacts of xenia and metaxenia, the chosen palms were pollinated in April using pollen grains from a male grown at the same place throughout the year. Three bunches, represented one of the chosen trees; the number of produced bunches multiplied by the bunch weight yielded the total yield per palm.

Yield parameters and fruit physical characteristics:

In October, with the full color (Rutb stage), samples of thirty fruits per replicate were collected to measure several physical characteristics, such as fruit and seed weight, flesh weight, percentage of flesh, and fruit dimensions.

Chemical properties:

- 1- A sample of roughly 50g of chopped flesh was taken and dried in a draft oven at 70°C until a consistent weight was achieved. The moisture content percentage was then calculated.
- 2- The Total soluble solids (T.S.S%) was determined by using a hand refractometer according to Verma *et al.* (2000). and Shamra and Nautiyal (2009).
- 3- Treatable acidity percentage in the flesh expressed as percentage by titration against Na OH (0.1 N) using phenol phthaline as an indicator, as described by A.O.A.C (2023).
- 4- Total phenols concentration was determined colourimetrically as described by Walid *et al.* (2022).
- 5- Total tannins concentration of date fruit peel was determined using the method described by Singleton *et al.* (1999).
- 6- Crude fibers determination was achieved by adding glacial acetic acid and nitric acid (10:1) on 1g fruit flesh samples as described by Marzieh *et al.* (2010) and A.O.A.C (2023).
- 7- Reducing and total sugars concentrations were determined according to Lane and Eynon method as described by Aseeri *et al.* (2021). The non-reducing concentration was calculated from the difference between total and reducing sugar.

Selection of semi-dry date palm genotypes:

The present selection was achieved using modified Abo Rekab (2005) method of numerical selection of the tested date palm genotype trees: One hundred points were used to determine whether tested date palm genotypes should be chosen. The average yield per palm (20 points) and the fruit quality attributes (80 units) Splitting the eighty points are total sugar (20 points), tannins (10 points), flesh weight (20 points), T.S.S. (15 units), total acidity (5 points), and crude fibers (10 points) were the distribution of the 80 points of fruit quality properties.

It has been proposed that the date palm tree with the highest value of any of the characters listed above be assigned the maximum points for selection in order to determine the numerical units of selection for yield, flesh weight, total sugar, and total soluble solids percentage. The

following formula would determine the equivalent number of points for the other date palm trees that were tested:

$$X = \frac{\text{Maximum units for character (x)} \times \text{Recorded value for character (x) of any tested date palm tree}}{\text{tree highest value for character (x)}}$$

On the other hand, for calculation of numerical units of selection for tannins, total acidity and crude fibers percentages; it was proposed that maximum units for any of the previous characters would be given to the date palm tree which had the lowest value of such character under study. The other tested date palm trees would be given comparable units according to the following formulation:

Maximum units for specific character (x) \times lowest value for character (x) / Recorded value for character (x) of any tested date palm tree.

ISSR-PCR analysis:

For ISSR-PCR testing, five primers were employed. PCR reactions were conducted using five arbitrary 10-mer primers. Their names and sequences are present and shown in **Table (1)**.

Table (1). List of primer names and their nucleotide sequences used in the study for ISSR procedure.

No	Name	Sequence
1.	HB11	5' ACC CGC AAG G 3'
2.	HB09	5' ACC CGC AAG G 3'
3.	HB13	5' TTG GCA CGG G 3'
4.	HB15	5' ACC CGC AAG G 3'
5.	HB08	5' AGG CAT CGT G 3'

For ISSR-PCR testing, five primers were employed. Amplification was put into practice contains the following reagents in a 25 µl/reaction volume: 1 µl of Taq polymerase (1 U/0l), 2.5 µl of 10 x buffer, 3.0 µl of primer (10 pmol), 3.0 µl of template DNA (25 ng/µl), 2.501 of dNTPs (2.5 µmol), and 2.5µl µg Cl₂ (2.5µmol). The PCRs were set up for 45 cycles, each consisting of 1 minute at 94 °C, 1 minute at 57 °C, and 2 minutes at 72 °C. The first cycle lasted 4 minutes at 94 °C. Ultimately, the reaction was held at 72°C for ten minutes. The 1.5% agarose gel was used to separate the PCR products, and the 100 bp ladder DNA marker (fermenters) with a sequence of 1000, 900, 800, 700, 600, 500, 400, 300, and 200 were used to estimate the fragment sizes.

Statistical Analysis:

For this study, a randomized complete block design was used, Snedecor and Cochran' (1980) conventional methods for statistical data analysis were applied. A new test called the L.S.D. was employed to compare the means. Each primer produced a count of DNA bands, whose molecular sizes were compared to those of the DNA markers.

Each primer's produced from DNA profile bands were combined into a single pool. Subsequently, every single DNA band was regarded as a binary character in a data matrix, with codes of 1 and 0, in order to determine genetic similarity and create a dendrogram tree based on the seven date palm plants under study.

RESULTS AND DISCUSSION

1- Yield parameters and fruit physical characteristics:

The **Table (1)** and **Fig. (1)** reveal that palm No. 3 generated the highest bunch weight, number of bunches, and total yield per palm when compared to the other semi-dry genotypes and the Sultany cultivar (Standard CV). The corresponding figures for bunch weight, quantity, and total yield per palm were 10, 7, 23, and 246.1 kg palm, respectively. The second season had the lowest bunch weight (10 kg), number of bunches (12/palm), and total yield (130 kg/palm) figures for the Sultany cultivar (control). The fruit weight and flesh weight figures for the two seasons under inquiry for the semi-dry seedy date palm are listed in **Table 1**.

The results showed that the highest ideals of the two characters were fulfilled. **Table (1)** displays the semi-dry genotype palms' fruit length and diameter values for the two seasons under investigation. The information gathered demonstrated that, when compared to other semi-dry genotype s, semi-dry palm genotype No. 1 produced the longest fruit, while palm No. 3 generated the largest fruit diameter. These findings were consistent with those of Hussein *et al.* (1979) and Badran and El-Shenawy (2009), who noted that pollen types affected the values of fruit length and diameter in different cultivars.

Table (1): Yield and its components (No. of bunches, bunch weight and total yield palm (kg) and fruit physical properties of the semi-dry seedy date palm selected under El-Wahat El-Baharia conditions during 2023 and 2024 seasons.

Palm genotypes No.	Fruit Weight		Flesh Weight		Fruit Length		Fruit Diam.		Seed Weight		No. of Bunches/ palm		Bunch weight/palm		Estimated yield / palm(kg)	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Sultany Cv.	7.44	8.34	6.17	7.03	4.03	4.00	2.07	2.03	1.28	1.31	12.0	13.0	10.5	10.0	126.0 ^e	130.0 ^e
T1	11.92	12.62	10.39	11.18	5.10	5.23	2.13	2.27	1.58	1.44	19.0	20.0	10.0	10.4	190.0 ^b	208.0 ^c
T2	10.29	9.39	8.26	7.58	4.13	4.33	2.32	2.30	2.03	1.81	18.0	20.0	10.1	10.5	181.8 ^c	210.0 ^c
T3	12.48	13.98	10.69	12.14	4.27	4.50	2.57	2.67	1.89	1.84	20.0	23.0	10.3	10.7	206.0 ^a	246.1 ^a
T4	9.58	10.89	8.01	9.35	3.93	4.03	2.33	2.40	1.57	1.54	19.0	21.0	10.2	10.6	193.8 ^b	222.6 ^b
T5	11.41	10.76	9.88	9.30	3.93	3.97	2.27	2.50	1.54	1.46	17.0	18.0	10.6	10.8	180.2 ^c	194.4 ^d
T6	11.60	12.58	10.11	10.93	3.40	3.67	2.50	2.53	1.49	1.66	18.0	19.0	10.0	10.5	180.0 ^c	199.5 ^d
New L.S.D.	1.72	1.96	1.49	1.76	0.29	0.16	0.28	0.26	0.44	0.46	1.36	1.41	0.02	0.01	6.85	7.01



Fig. (1): The fruit of a particular six genotype of semi-dry date palm and sultany.

2- Chemical properties:

The Total soluble solids (TSS %):

Regarding to TSS (%) it is clear that fruits of palm No. 3 significantly contained in the first and second seasons the highest T.S.S% values (57.1 and 58.5) as compared to the other values of semi-dry seedy palms. Meanwhile, palm of the control gave the lowest values (38.2 and 42, respectively). Similar trends in total soluble solids (TSS) were also noted, where palm genotype T3 exhibited the highest TSS values in both seasons, corroborating the findings by Awad (2006) and Badran and El-Shenawy (2009) regarding the influence of female variety on

TSS levels. These results are in conformity with those reported by Awad (2006) and Badran and El-Shenawy (2009) who reported that TSS % directly influenced by female variety.

Total Acidity (%):

The overall acidity percentage for the Sultany cultivar (the control) ranged from 0.33% to 0.16%, according to the semi-dry seedy date palm data. However, a comparison of palm No. 3 and No. 4 with the other genotypes under study showed noticeably lower overall acidity percentage values. Similar results were also observed by Badran and El-Shenawy (2009) and Baker *et al.* (2003).

Total, sugars, reducing and non-reducing sugars (%) contents:

The overall sugar proportion is both dropping and decreasing. In comparison to the other seedy palm values, palm No. 3 provided the highest total and non-reducing sugars (%) values with the same line of the TSS% in the semi-dry date palm seedy data (**Table 2**). **Table (2)** makes it evident that there were no discernible variations in the percentage of reducing sugar among the genotypes under investigation. However, when compared to the other genotypes under study, the Sultana cultivar displayed a noticeably lower value in both seasons. Similar findings were found in this regard by Hassan (2008), who claimed that female variety directly affected total sugars, and Al-Shaikh (2006); who reported that the effect to both cultivar and year.

Tannins %:

The data in **Table (2)** show that, in comparison to the other genotypes of trees under investigation, palm tree no. 3 produced the lowest tannin values in both seasons. Conversely, Sultany, the control cultivar, showed the highest tannin values. Scholars Abdalla (2002) and Abo-Rekab (2005) reached comparable findings. The results showed that palm No. 6 had the lowest value of fibers % during the first season, whereas the highest values of fibers % during the two seasons were obtained by the control (Sultany cv). These results were in accordance with those obtained by Abo-Rekab (2005).

Indoles (%):

Table (2) data demonstrated that the semi-dry genotype date palm No. 3 indoles concentrations had the highest values through the study's two seasons, producing 0.134 and 0.140%, respectively. Abo-Rekab (2005) achieved similar results.

Phenols (%):

The data tabulated in **Table (2)** makes it evident that, in both research seasons, the genotype No. 1 semi-dry date palm produced the lowest total phenols percentage when compared to the other values of the other genotypes. The control (Sultany cv.) produced the highest values (0.554 and 0.583%) in both seasons.

Moisture content (%):

The data presented in **Table (2)** highlights that moisture content varied significantly among the different semi-dry date palm genotypes. Specifically, genotype T4 displayed the highest moisture content during both seasons, indicating a notable difference in comparison to other genotypes. This trend suggests that moisture retention is genotype-dependent, with Sultany Cv. (control) showing the lowest values at 22.2% and 23.33% for the first and second seasons, respectively. These findings are consistent with the research by Al-Saikh (2006), who documented similar moisture content variations across different date palm varieties. Moisture content is critical in determining the shelf life and quality of dates, as it influences

the texture, microbial growth, and overall storage stability. Increased moisture levels, as observed in genotype T4, can lead to faster spoilage, whereas lower moisture percentages, as seen in the control group, may contribute to better preservation over time.

Table (2): Comparative analysis of physicochemical properties of semi-dry date palm genotypes under El-Wahat El-Baharia conditions during 2023 and 2024 seasons.

Palm genotypes No.	TSS (%)		Total acidity (%)		Total sugar (%)		Reducing sugar (%)		Non-reducing sugar (%)		Tannins (%)		Indoles (%)		Phenols (%)		Fibers (%)		Moisture (%)	
	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2	S1	S2
Sultany Cv.	38.2	42.0	0.3	0.3	68.2	72.1	31.3	33.4	36.9	38.7	0.4	0.4	0.1	0.1	0.6	0.6	3.5	3.6	22.2	23.3
T1	49.4	52.7	0.2	0.2	79.4	80.5	35.0	35.2	44.4	45.3	0.1	0.1	0.1	0.1	0.1	0.1	3.0	3.1	26.0	26.3
T2	48.0	49.4	0.3	0.3	78.1	79.5	35.9	36.3	42.2	43.2	0.1	0.1	0.1	0.1	0.2	0.3	3.2	3.2	28.1	28.6
T3	57.1	58.5	0.2	0.2	80.6	82.0	34.5	35.8	46.1	46.2	0.1	0.1	0.1	0.1	0.2	0.2	3.3	3.4	25.4	25.6
T4	51.2	56.7	0.2	0.2	79.2	80.3	36.1	34.0	43.2	46.3	0.2	0.2	0.1	0.1	0.3	0.3	3.1	3.3	29.2	29.5
T5	46.6	49.2	0.3	0.3	76.1	78.6	35.7	35.8	40.3	42.8	0.2	0.2	0.1	0.1	0.3	0.3	2.9	3.1	27.2	27.8
T6	46.9	52.2	0.3	0.3	77.6	79.6	36.3	36.7	41.3	42.9	0.2	0.2	0.1	0.1	0.3	0.3	2.7	3.3	24.4	24.9
New L.S.D.	1.8	2.2	0.0	0.0	1.7	1.2	1.2	1.5	15.0	1.6	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.4	1.78	1.72

General evaluation:

Table (3) summarizes the general assessment of the Sultany cultivar (control) and the selected genotypes of semi-dry date palms. Compared to the other semi-dry genotype palms, the semi-dry date palm No. 3 achieved the largest accumulative units (97.66 units), ranking first in the general evaluation for productivity and fruit quality. The second was given by Palm No. 1, which achieved (91.27 units). The semi-dry date palm genotype No. 4 ranked the third place with 86.78 units, and tree No. 6 obtained the fourth rank with 84.4 units. Genotype tree No. 2 achieved the fifth rank, scoring 82.68 units. With 81.46 units, the semi-dry genotype of date palm No. 5 achieved the sixth rank. The data in **Table (3)** evaluates several parameters of an overall palm including yield per palm, flesh weight, total sugars, total soluble solids (TSS), total acidity, tannins and crude fibers. The findings revealed high variation between the two genotypes and firmly support the idea that the quality and yield of the fruits are sharply genotype dependent, and the genetic contents of each genotype which independently regulate the quantity and quality of the fruits.

Yield per palm (kg):

In terms of yield, genotype T3 recorded the highest yield with an average of 20.00 kg per palm; T4 genotype yield recorded an average of 18.40kg. This is a slight improvement when compared with control (Sultany Cv.) which produced 10.56 kg. Enhanced yields can be attributed to the closer adaptation of these genotypes to environmental conditions, from genetic merit and enhanced best practice. Al-Hooti *et al.* (1997) also arrived a similar conclusion of the genetically induced variability in the date palm yield potential.

Flesh weight and total sugars (%):

T3 had highest flesh weight and maximum total sugars reaching of 20.00 index units. Also, another main factor of making the dates ordinarily acceptable by the market or preferred by the consumer since it affects the quality of the edible portion. Higher aggregate concentration of total sugar in T3 not only has a sweet taste where it is preferred for consumption. These findings differ with those of Ahmed and Ahmed (2005) who showed that

higher flesh weight and sugar content genotypes have more consumer preference and higher economic value.

Total soluble solids (TSS) and total Acidity:

Total soluble solids (TSS) tend to be used as an index of fruit maturity and their sweetness. From the result s of the TSS analysis, the TSS index score the highest at 15.00 units for the palm genotype T3 and the control had the lowest score of 10.77 units. TSS was positively related to fruit sweetness and taste, resulting in a better genotype for fresh markets or for processing. Also, Al-Farsi *et al.* (2005) opined that in date palms, TSS levels did differ between genotypes and were significant contributor to palace in fruits. However, total acidity was significantly higher in T4 depicting an average of 5.00 index units which can be suggestive of higher sourness compared to the other genotypes. A pH reduces the overall quality of dates and sweetness and relates to the sweetness and the pH values, the willingness of the consumers and the processing characteristics.

Tannins and crude fibers:

Tannins are valuable due to their being antioxidants and their input towards the astringency of dates. Genotype T3 contained the highest tannin content at 10.00 index units hence the genotype has potential of being a source of antioxidants, which are beneficial to the consumer. On the other hand, T6 had the highest crude fiber content of 10.00 index units; a factor that enhances the nutritional fiber in dates. High crude fiber is essential for digestion and also has a nutritional value for the body. Habib and Ibrahim (2011) have also identified the same effect reflecting that tannin and fiber levels in dates add value to functional values of dates properties to the dates.

General evaluation of palm:

Taking an overall view of the results, genotype T3 was the highest yielding with 97.66 units while genotype T1 scored 91.27 units. These outcomes show that those specific genotypes give better quality and yield traits making them more fit for cultivation and marketing. The control (Sultany Cv.) has the least overall general evaluation of 63.56 units and therefore has been considered as less effective in terms of yield, quality and performance as compared to the selected commercial brand. These observations conform with other investigations by Barreveld (1993) who supported the concept of phenotypic assessment of various desirable and undesirable traits are the sole purpose of deciding the most productive genotype of date palm for either fresh purpose, dry purpose or processing.

Table (3): Comprehensive Evaluation of Semi-Dry Date Palm Genotypes.

Character		Yield/palm (kg)	Flesh weight	Total sugars (%)	T.S.S. (%)	Total acidity (%)	Tannins	Crude fibers (%)	General evaluation of palm
Palm genotype	Index units specified	20	20	20	15	5	10	10	100 units
Sultany Cv.		10.56	11.58	17.50	10.77	2.50	2.72	7.93	63.56
T1		16.90	18.42	19.63	13.51	3.81	9.73	9.27	91.27
T2		17.07	13.61	19.39	12.67	2.96	8.12	8.86	82.68
T3		20.00	20.00	20.00	15.00	4.21	10.00	8.45	97.66
T4		18.40	15.40	19.59	14.54	5.00	4.82	9.03	86.78
T5		15.80	16.28	19.17	12.62	2.86	5.00	9.73	81.46
T6		16.21	18.00	19.41	13.38	2.16	5.24	10.00	84.40

Identification of ISSR-PCR markers:

Randomly repeating short DNA sequences served as the foundation for the ISSR class of molecular markers. Because these non-functioning sections lacked functional constraints, even amongst genotypes that were closely related, these repetitions were highly variable. Similarly, a substantial degree of polymorphism was found with the two ISSR-PCR primers.

Tables (4–8), Thirty ISSR bands were collected in total. Out of these thirty bands, sixteen (53.33%) were monomorphic and fourteen (46.67%) were polymorphic. The greatest quantity of amplifications was produced. The levels of polymorphism varied amongst primers. The polymorphisms of primers HB-08 (62.5%) and HB-15 (66.67%) were both high. Conversely, primers HB-09 (33.33%) and HB-13 (40%) showed moderate expression levels. Primer HB-11 showed just 20% polymorphism.

Table (9) displays the total number of amplified fragments, polymorphic bands, monomorphic bands, and unique bands for each primer of each genotype and Sultany cultivar utilizing the five primers. Each semi-dry date palm genotype and Sultany cultivar was distinguished from the others by the following particular fragments:

Three distinct fragments were identified by primer HB-08; two of these served as negative markers for genotype 2 and only one as a positive marker for genotype 6. Two distinct fragments were identified by primer HB-09 as one positive marker for genotype 4 and one negative marker for genotype 6. Primer HB-11 failed to identify any particular pieces. There was only one negative marker displayed by both primers, HB-13 and HB-15, for each **Fig (2)**.

The genetic similarity between the six genotypes of semi-dry date palms and the sultany cultivar was estimated using the ISSR data and UPGMA computer analysis, as shown in **Table 10**. Between semi-dry date palms Nos. 2 and 4, the similarity index reached the highest (1.0), but between Nos. 3 and 5, the similarity index was lowest (0.0). Figure 3 depicts a dendrogram that shows the genetic relationships between the six semi-dry date palm genotypes, dividing the genotypes into two main clusters along with the Sultany cultivar. The semi-dry palm No. 2 was the lone member of the first cluster. The second cluster was further divided into two sub-clusters, the first of which contained the semi-dry date palm No. 6.

The Tables (4 to 8) fulfilled the results presented in these tables. Genetic Differences for Genotypes of the studied Date Palm (**Tables, 4:8**).

PCR-RFLP analysis with ISSR primers (HB-08, HB-09, HB-11, HB-13, and HB-15) displays the percentage polymorphism in the six semi-dry date palm genotypes and the Sultany cultivar. Polymorphism is essential because variation within DNA is essential for adaptation, diseases resistance, and other applications of breeding programs.

HB-08 Primer (Table 4):

The HB-08 primer generated a total of 8, of those, only five loci were found to be polymorphic in our study population set, which gives us a polymorphism rate of 62.50%. As indicated by the fact of unique bands (three), it can be assumed that some genotypes are endowed with specific genes, for instance, T4 and T6. This relatively high level of polymorphism means that this primer may be useful for discrimination among genotypes. (Al-Qurashi & Awad, 2011).

HB-09 Primer (Table 5):

The total numbers of bands produced by the HB- 09 primer were six but there were only two polymorphic bands and the polymorphic percentage was found to be 33.33%. These show that this primer did not detect a higher genetic diversity than that detected by HB-08.

However, two bands of specific length were observed, thus underlining the usefulness of this primer in identifying specific mutation in some genotypes. (Elmeer & Almalki, 2011).

HB-11 Primer (Table 6):

HB-11 primer yielded five bands in all, of which only one was polymorphic giving a polymorphism percentage of 20%. This is the lowest polymorphism that has been observed among the primers and indicates that perhaps HB-11 will not be as effective for the identification of genetic variation between the genotypes. No specific bands were observed on using this primer. (Hamwieh *et al.*, 2009).

HB-13 Primer (Table 7):

HB-13 primer showed polymorphic five bands out of which two are polymorphic, so the polymorphic percentage is 40%. First, there is one band which suggests genotype dependent characteristics. The moderate polymorphism indicates that this primer ought to classify among the genotypes with a passable amount of success. (Al-Qurashi & Awad, 2011).

HB-15 Primer (Table 8):

The HB-15 primer produced six total bands in total with 4 of them being polymorphic with what lead to the highest polymorphism percentage of 66.67 percent. This primer was particularly efficient in identifying genetic differences because a large number of polymorphic and unique bands. Thus, the degree of genetic variability evidenced by HB-15 could be said to point to the fact that the latter is a veritable weapon for distinguishing between the genotypes of date palm. (Elmeer & Almalki, 2011).

Relationships genetic:

The ISSR Dice (1945) coefficient genetic similarity matrix illustrates the parameters of the genetic dissimilarities between six semi-dry date palm genotypes as well as Sultany cultivar. A higher value of similarity coefficient tells the two genotypes are more related in a genetic point of view, while lower value indicates that they are genetically more-distant.

When the study's results were analyzed, it was found out that Genotype 3 and Genotype 4 had a genetic similarity of one, meaning that they are the closest relatives genetically either being of the same genotype or at least belong to the same genotype set. This could be because these two genotypes may share a common origin or there is little variation among the genotypes. (Al-Qurashi & Awad, 2011).

In the present study, the similarity coefficient between Sultany cultivar (control) and Genotype 1 was 0.28 while for Genotype 2 it was 0.52, which indicated that there is moderate genetic variability available in the present semi-dry date palm genotypes. (Hamwieh *et al.*, 2009).

C4 was shown to have the largest genetic distances from other genotypes, especially with C5 genotype = 0.0, which categorized C4 and C5 as distinct genetic strains. This may be useful for the development programs, because the introduction of genetically diversified material could contribute to the improvement of the further varieties of date palm. (Elmeer & Almalki, 2011).

The data obtained in the current study show that there is a genetic differentiation between the six semi-dry date palm genotypes and the Sultany cultivar as identified using the ISSR. The polymorphic percentages are higher in HB- 08 and HB-15, which show higher genetic diversities essential for choosing the highest yielding genotypes for breeding and cultivation. The ISSR markers were useful for the identification of different genotypes that may be harnessed for breeding efforts targeting yield, disease and pest resistance and response to changing environmental conditions.

This is evident due to high genetic relationship between different genotypes; for instance, Genotype 3 and Genotype 4 which might have originated from a similar genotype type or have faced similar genetic pressures. On the other hand, the existence of considerable genetic distinctiveness of Genotype 4 from other genotypes, including genotype 5, offer chances to bring new genetic characteristics in to breeding programmes for genetic improvement and perpetuity of the breed.

Finally, the results obtained with different primers show that ISSR can successfully be used to analyze genetic polymorphism of semi-dry date palm genotypes. The fact that the level of polymorphism differed so much between the loci indicates that some primers, such as HB-15, allow for the identification of genetic differences. Thus, the estimates of genetic similarity between contributors to the next generation are also useful for breeding programs to identify how genotypes should be interbred so as to maximize the dissimilarity improve desired traits.

Table (4): Survey of ISSR using HB-08 primer in six semi-dry date palm genotypes and one Sultany cultivar (S). (1) Means presence and (0) means absence.

Band No.	M.W bp	1	S	2	Date Palm Cultivars 3	5	6
	940		0	0			
	800		0	0		0	
	760			0			
	600						
5	450						
6	130	1					
	230			1			
	160	0		0			
Total		6	6	3	7	7	8

Table (5). The HB-09 primer is used to survey the ISSR in six semi-dry date palm genotypes and one Sultany cultivar (S); the characters (1) and (0) denote presence and absence, etc.

Band No.	M.W bp	Date Palm Cultivars						
		1	S	2	3	4	5	6
1	580							0
2	430							
3	300	1		1	1			
4	240	1	1	1	1			1
5	170	1	1	1	1		1	1
6	100	0	0	0	0	1	0	0
Total		5	5	5	5	6	5	4

Table (6): Shows the results of an ISSR assessment using the HB-11 primer in six semi-dry date palm genotypes and one Sultany cultivar (S); the letters (1) and (0) represent presence and lack, respectively.

Band No.	Date Palm Cultivars							
	M.W bp	1	S	2	3	4	5	6
1	480	1	1	1	1	1	1	1
2	460	1	1	1	1	1	1	1
3	360	0	0	0	1	1	1	0
4	280	1	1	1	1	1	1	1
5	180	1	1	1	1	1	1	1
Total		4	4	4	5	5	5	4

Table 7: HB-13 primer survey of six semi-dry date palm genotypes and one Sultany cultivar (S); (1) denotes presence, while (0) denotes lack.

Band No.	M.W Bp	Date Palm Cultivars						
		1	S	2	3	4	5	6
1	640	0	0	0	1		1	0
2	540	1	1	1	1	1	1	1
3	480	1	1	1	1	1	1	1
4	370	1	1	1	1	1	1	1
5	340	1	1	1	1	0	1	1
Total		4	4	4	5	3	5	4

Table 8: ISSR survey with HB-15 primer in six semi-dry date palm genotypes and one Sultany cultivar (S); letters (1) and (0) denote presence and absence, respectively.

Band No.	M.W Bp	Date Palm Cultivars						
		1	S	2	3	4	5	6
1	820	0	1	0	1	1	1	1
2	500	1	1	1	1	1	1	1
3	430	1	1	1	1	0	1	0
4	345	0	1	0	1	1	0	0
5	220	1	1	0	1	1	1	1
6	185	1	1	1	1	1	1	1
Total		4	6	3	6	5	5	4

Table (9): The total number of bands matched with the polymorphic, unique, monomorphic, and polymorphic percentages is provided as follows:

Primer name	Total band	Polymorphic band	Monomorphic band	Unique band	Polymorphic %
HB-08	8	5	3	3	62.50
HB-09	6	2	4	2	33.33
HB-11	5	1	4	-	20.00
HB-13	5	2	3	1	40.00
HB-15	6	4	2	1	66.67

Table (10): Shows the genetic similarity matrices generated with the ISSR dice coefficient for the six semi-dry genotypes and Sultany cultivar (S) accessions.

.	1	S	2	3	4	5
S	0.28					
2	0.34	0.52				
3	0.35	0.17	0.76			
4	0.55	0.35	1.0	0.24		
5	0.27	0.26	0.68	0.0	0.33	
6	0.39	0.37	0.84	0.43	0.45	0.35

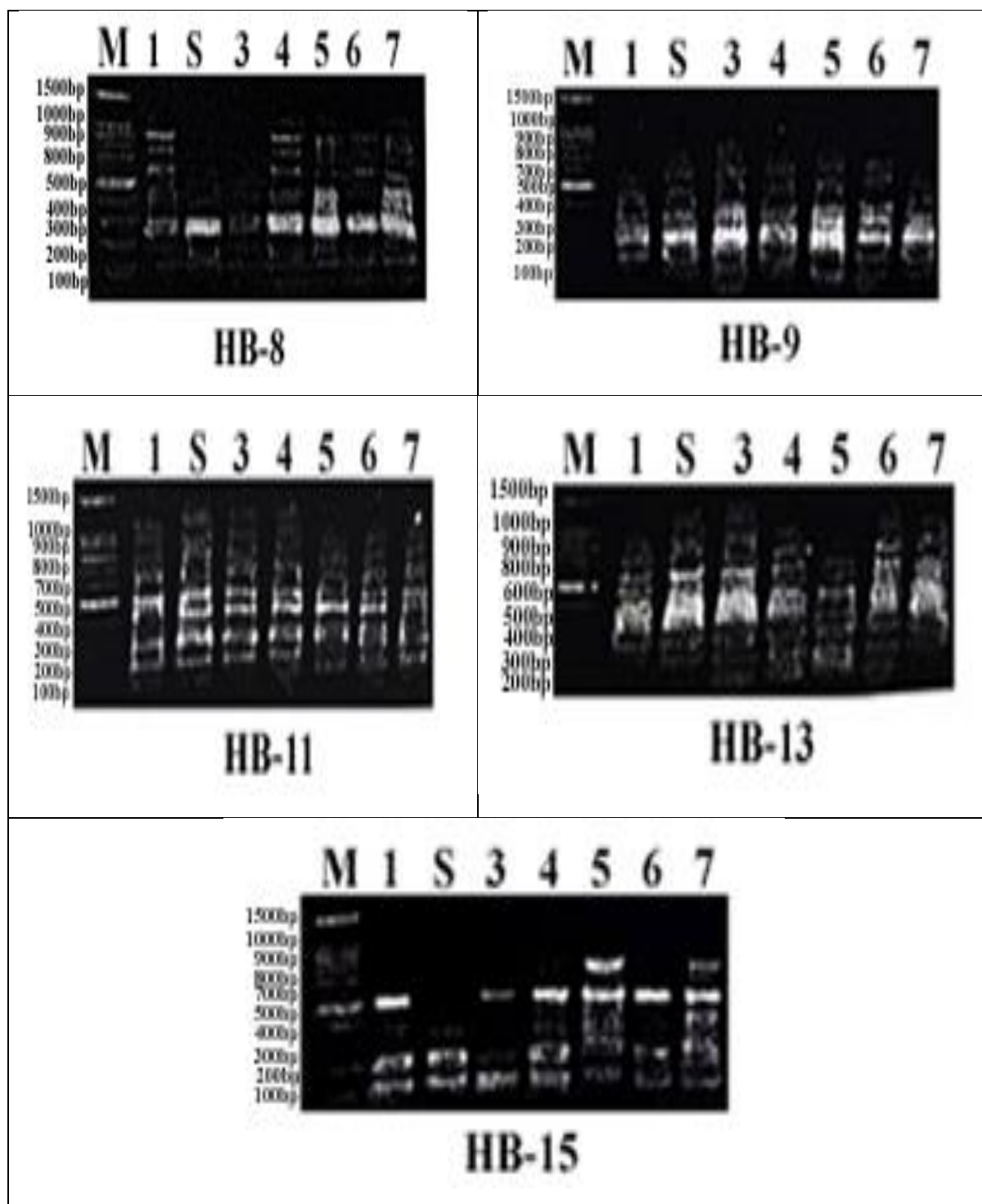


Fig. (2): ISSR products made with primers HB-08, HB-09, HB-11, HB-13, and HB-15 from six genotypes of semi-dry date palms and one Sultany cultivar.

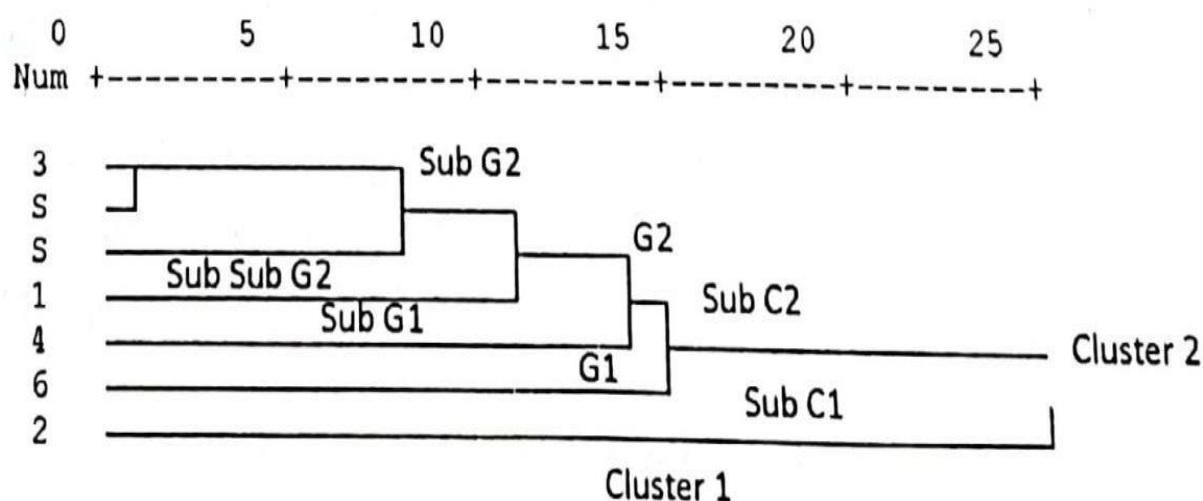


Fig. (3): Using similarity matrices built with the dice coefficient and unweighted pair-group arithmetic (UPGMA data), dendrograms for the six semi-dry date palm genotypes and the Sultany cultivar (S) accessions have been developed from the ISSR data.

Conclusion

It was possible to reliable selection of superior semi-dry date palm putative genotypes in El-Wahat El-Dakhla, New Valley Governorate that may be used for production of high-quality fruits. Therefore, the study results for the two seasons (2023-2024) showed that some putative genotypes such as genotype number 3 recorded better yield traits compared to the Sultany cultivar used as a control. The study on physical evaluation of fruits showed that apart from big size, selected semi-dry genotypes possessed better length and diameter, which suggest the possibility of commercial viability of fetching high amount for the crop. Besides, use of ISSR-PCR analysis gave the molecular view of the present work which supports the studies on genetic variability among the concerned genotypes. The systematic assessment of the examined palm trees and the integration of the phenotypic and molecular data on the selected genotypes highlight these palm trees as prospective for further cultivation with a view to increasing the production of dates in the affected areas. This study could serve as the basis for adopting superior date palm genotypes to the Dakhla Oasis environment, which will result in the sustainable use of resources and the enhancement of the economy of date palm production in the New Valley area of Egypt.

REFERENCES

- A.O.A.C. (2023). Official Methods of Analysis 18. Association of official Agriculture Chemists. 12 Ed. Published by A.O.A.C Washington D.C. USA.
- Abdalla, A.S. (2002). Evaluation of three date palm genotype types grown at El-Dakhla oasis New Valley, Egypt. Minia Journal of Agricultural Research, volume 22 No. 2 (c) 2002, Special Issue (The proceedings of Minia 1st Conference for Agricultural and Environmental Sciences (MCAES 15) March 25-28, Minia-Egypt).
- Abdallah, O.S., Zeroual, I.D., Randa, G., Said, G. (2020). *In vitro* and Molecular Docking Studies of DPPH with *Phoenix dactylifera* L. (Deglet-Nour) Crude Fruits extracts and Evaluation of their Antioxidant Activity 13(1): January -February 2020: 52-59.

- Abo-Rekab, Zeinab A.M. (2005). Some physiological studies on date palm. Ph. D. Thesis, Faculty of Agriculture, Cairo University, Egypt.
- Al-Farsi, M., Alasalvar, C., Morris, A., Baron, M., Shahidi, F. (2005). Compositional and sensory characteristics of three native sun-dried date (*Phoenix dactylifera* L.) varieties grown in Oman. *Journal of Agricultural and Food Chemistry*. 53(19): 7586-7591.
- Al-Hooti, S., Sidhu, J.S., Al-Otaibi, J. (1997). Chemical composition and quality of date syrup as affected by pectinase/cellulase enzyme treatment. *Food Chemistry*. 59(1):153-157.
- Al-Qurashi, A.D., Awad, M.A. (2011). Genetic diversity among date palm (*Phoenix dactylifera* L.) genotypes using ISSR markers. *Journal of Horticultural Science and Biotechnology*, 86(3), 283-287.
- Al-Shaikhan, M.S. (2006). Physical and chemical characteristics response of three date palm cultivars to source of pollen grains. *J. Agric. Sci. Mansoura Univ.*, 31 (3): 1537-1546.
- Aseeri, I.A., Omar, A.K., Shareef, H., Aly, J.K. (2021). Clean agriculture for the safe production of date palm fruit (*Phoenix dactylifera* L. cv. Sewi) under Egtptian conditions. *Applied Ecology and Environmental Research* 19 (5):3551 -3561. <http://www.aloki.hu> (Online) DOI: http://dx.doi.org/10.15666/aer/1905_35513561, Budapest, Hungary.
- Awad, M.A. (2006). Fruit set failure and abnormal fruiting of tissue culture derived "Nabout Saif" date palm as affected by pollinator type and pollination intensity. Third International Date Palm Conference, Abu-Dhabi, United Arab Emirates, 19-21 February.
- Barreveld, W.H. (1993). Date Palm Products. FAO Agricultural Services Bulletin No. 101. Rome, Italy: FAO.
- Badran, M.A.F., El-Shenawy, M.A. (2009). Evaluation of some soft and semi-dry date palm genotype clones growing under Assiut conditions. *Mnia J. of Agric. Res.; Develop.*, 29 (2) :317-331.
- Baker, E.I.P., Haseeb, G.M., Metwally, A.A.M. (2003). Selection of a superior semi-dry date palm tree under Assuit (Egypt) conditions. *Proc. Of the International Conference on date palm*, 16-19 September in Saudi Arabia, 759-780.
- Dice, L.R. (1945). Measures of the amount of ecologic association between species. *Ecology*, 26:297-302.
- Elmeer, K., Almalki, S. (2011). Use of inter simple sequence repeat markers to detect genetic diversity of date palm (*Phoenix dactylifera* L.) in Qatar. *Agriculture and Biology Journal of North America*, 2(3):314-320.
- Habib, H.M., Ibrahim, W.H. (2011). Nutritional quality of 18 date fruit varieties. *International Journal of Food Sciences and Nutrition*, 62(5):544-551.
- Hamwieh, A., Udupa, S.M., Sarker, A., Baum, M. (2009). Development of new microsatellite markers and their application for genotyping in lentil (*Lens culinaris* Medik.). *Euphytica*, 168(3), 457-469.
- Hassan, A.R. (2008). Physiological studies on some date palm under Assuit conditions. Ph. D. Thesis, Hort. Dept. Agric. Fac., Assuit University.
- Hussein, F., Al-Kahtani, M.S., Wally, Y. A. (1979). Date palm growing and date production in the Arab and Islamic world. Ain Shams Univ. Press (In Arabic).
- Marzieh, S., Keikhosro, K., Mohammad, J.T. (2010). Palm Date Fibers: Analysis and Enzymatic Hydrolysis *Int. J. Mol. Sci.* 2010, 11, 4285-4296.

- Mostafa, A.A., El-Aidy, A.A., El-Sammak, A.P. (1986). Evaluation and study of some genotype date palms grown at El-Wady El-Gedid. Second Symposium on Date Palm. March 3- 6. King Faisal Univ., Saudi Arabia pp. 153-161.
- Popenoe, P. (1973). The date palm. Field Research Projects Coconut Grove, Miami, Florida.
- Samir, Z., Ismail, D., Gaouaoui, Randa, Said, G. (2020). *In vitro* and Molecular docking studies of DPPH with (*Phoenix dactylifera* L.) Deglet-Nour crude fruits extracts and Evaluation of their Antioxidant Activity. Asian J. Research Chem. 13(1): January - February 52-58.
- Shamra, S.K., Nautiyal, M.C. (2009). Post Harvest Technology of Horticulture Crops. Published by New Indian Publishing Agency, Pitampura, New Delhi-110 088. Pp. 111-113.
- Singleton, V.L., Orthofer, R., Lamuela Raventós, R.M. (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Methods in enzymology, 299,152-178.
- Snedecor, G.W., Cochran, W.G. (1980). Statistical Methods. Oxford and J.B.H. Publishing Comm., 6th Edition.
- Soliman Kh., Rizk, R.M., El-Sharabasy, S.F. (2006). Genetic polymorphism of semi-dry date palm (*Phoneix dactylifera* L.) cultivars in Egypt. J. Biotechnology. 22, 261-273.
- Swain, T., Hills, W.E. (1959). The phenolic constituents of *Pruns domestica*. 1-The quantitative analysis of phenolic constituents. J. Sci. Food. Agri., 10:63-68.
- Verma, L.R., Joshi, V.K. (2000). Quality assurance for fruits, vegetable and their products. In: Post harvest technology of fruits and vegetables. Tata McGraw-Hill. Publishing Company Limited, New Delhi, India. 2000 - 1240.
- Walid, M.K., Ruba Abuamsha, Nisreen Alqaddi (2022). Phenotypic Characterization of Local Date Palm Cultivars at Jericho in Palestinian Jordan Valley District Indian Journal of Science and Technology 2022;15(20):989–999.
- Zehdi, S.L., Sakka, H.L., Rhouma, Ould A., Mohamed, S., Marrakchi, M.L., Trifi, M.L. (2004). Analysis of Tunisian date palm germplasm using simple sequence repeat primers. African J. of Biotech., 3(4): 215-219.