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Evaluation of phenotypic relationships of date palm cultivars at Melka Werer, Ethiopia

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ABSTRACT: Date palm (*Phoenix dactylifera* L.) is one of the oldest fruit bearing perennial trees classified under the genus of *Phoenix* in Arecaceae (Palm) family. The natures of date palms with their long productivity have an indispensable socio-economicsignificance and contributions in agricultural sustainability. The date palm is a food and income source for societies particularly in the desert and also it has medicinal, cultural, ecological and environmental values. Date palms are dioecious with a wide range of phenotypic variations. Therefore, evaluation of date palm cultivars using morphological characters at regional as well as global level is critically necessary for sustainable utilization of its genetic resources as well as for genetic improvement and conservation programs. The aim of this study is to assess the phylogenetic relationships of ate palms date palm cultivars cultivated at Melka Werer research centre. A total of 45 morphological traits were used to assess phylogenetic relationships of eleven date palm cultivars. All traits showed mean variations among the cultivars. Principal component analysis on the first component revealed 37% variation in vegetative and reproductive traits data combined together and 29% and 32% of variations was observed in separate vegetative and reproductive morphological traits data respectively. Among 45 morphological traits 43 traits exhibited significant differences at $p < 0.05$ in analysis of variance and also 23 traits showed significant variances at $p < 0.05$ in homogeneity of variance analysis among cultivars. Besides, dendrograms were constructed based on combined vegetative and reproductive traits data and in separate vegetative and reproductive traits data and showed the genetic relationships between date palm cultivars. Generally, this study clearly display the phenotypic variations between cultivars and also the result is important as baseline for documenting and further agronomic traits studies of date palm cultivars particularly in Ethiopia.

Key words/phrases: ANOVA, Cultivars, Morphological traits, *Phoenix dactylifera* L., Principal component

INTRODUCTION

Date palm (*Phoenix dactylifera* L.) is a monocotyledon dioecious perennial tree that belongs to Arecaceae (Palms) family under the genus of *Phoenix*. It is one of the oldest fruits bearing cultivated crop with its long productivity that grows in arid and semi-arid part of the world. This crop has long history of cultivation and germplasm exchange in North Africa and Middle East countries and its origin is believed to be in

Mesopotamia (Chao *et al.*, 2007). Date palms have great socio-economic impact and an eminent contribution in agricultural sustainability (Al-Khalifah and Shanavaskhan, 2012). It is a multi-purpose tree having food, industrial, commercial, medicinal and ornamental values (Johnson, 2012; El-Far *et al.*, 2016). Date fruits have high nutritional value and contain about 70% sugar, essential vitamins, minerals and different value-added products are produced (Al-Khalifah and Shanavaskhan, 2012). Different parts of date palm are used for different purposes (Fatima *et al.*, 2016);

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trunks to construct houses, hives, bridges and as packing material (Al-Jabri, 2014); terminal buds and young leaves can be cooked as vegetables while rachises are used for paper making (El-Hadrami and Al-Khayri 2012; Khiari *et al.*, 2011).

In Ethiopia, date palm cultivation started approximately 200 years ago in Afrar region at Afambo and Asayta and other places which borders to Awash River; it was introduced to Errer Gota and Dire Dawa areas by Arabian trader who settled on these areas. Hussien, (2010) and Ben-Salah (2015) pointed out Afars have established date farms in areas around the Awash delta and Afambo region of Ethiopia. For adaptation and improvement programme of date palms in Ethiopia 14 *in vitro* date palm varieties have been introduced and planted at Afambo (Humodoyta site), Asayta and Melka Werer (Melka Werer Agricultural Research Centre). Perennial crops including date palms have diverse life history and breeding systems unlike annual crops. Morphologically, date palm cultivars are highly diversified in both vegetative and reproductive traits. In date palms phenotypic traits, especially, the reproductive parts have great role to discriminate degree of polymorphism between cultivars (Simozrag *et al.*, 2016; Bedjaoui and Benbouza, 2018). Assessment of diversity and phylogenetic relation of date palm cultivars using morphological data have been reported in Tunisia (Hamza *et al.*, 2011; Kadri *et al.*, 2019; Karim *et al.*, 2021; Ouardaet *et al.*, 2012), Algeria (Simozrag *et al.*, 2016; Bedjaoui and Benbouza, 2018; Abdelkrim *et al.*, 2020), Iraq (Khierallah and Azhar, 2016), Egypt (Eissa *et al.*, 2009; Ibrahim *et al.*, 2014; El-Sharabasy and Rizk, 2005), Sudan (Elsafy *et al.*, 2015), Pakistan (Haider *et al.*, 2015; Faqir *et al.*, 2018), Iran (Khankahdani and Bagheri, 2019), Saudi Arabia (Al-Khalifah *et al.*, 2012), Morocco (Elhoumaizi *et al.*, 2002) and Nigeria (Odewale *et al.*, 2013). Date palm cultivars exhibit a wide range of phenotypic variations due to their out-breed nature as well as dates is as one of staple fruit that easily distributed commercially throughout the world. These factors enhance the evolutionary diversification and

adaptability of palms out of native area. Hence, for sustainable utilization of genetic resources of date palm, their genetic improvement particularly related to agronomic traits and for introduction of date palm germplasm exploitation effective identification of date palm cultivars is critically important at regional and global level. Therefore, the objective of the present study is to assess the phylogenetic relationship of early introduced date palm cultivars using morphological traits that are cultivated at Melka Werer research centre, Ethiopia.

Materials and methods

Study area description and morphological data collection

Morphological study for eleven date palms cultivars was conducted using 45 traits (Table 1) at Melka Werer Agricultural Research Centre which is located at 10°9'59N latitude and 40°8'43E longitude in Amibara district of Afar Regional State of Ethiopia. The altitude of this area is 560 meters above sea level and the climate is hot and dry and the mean annual temperature of this area is 29.5°C. The study cultivars were recently introduced from England and Israel and planted at the same time and equally irrigated from Awash River throughout a year. These date palm cultivars are namely: Saggii, Khalas, Berhee, Ashal Al Hassa, Madjool England, Medjool Israel, Zamlli, Shishi, Khyara, Khadrawy and Jarvis (male). These cultivars were planted with 10 meters gaps between rows as well as palm trees. Morphological data using their vegetative and reproductive characters were collected from 11 date palm cultivars and each cultivar has four replications (4 trees used). One frond per tree and four fronds per cultivar and a total of 44 fronds were used to measure leaf traits; three fruits per tree and 12 fruits per cultivar and a total of 120 fruits from 10 cultivars (females) were collected to study phenotypic variation of date fruits.

Table 1. Morphological traits and their codes that were used for analysis.

Vegetative traits	Code	Unit	Reproductive traits	Code	Unit
Trunk height	T1	cm	Middle spine width	T24	Cm
Trunk circumference	T2	cm	Middle spine length	T25	Cm
Fronde length	T3	cm	Peduncle length from its base to the first spikelet	T26	Cm
Fronde width at the middle	T4	cm	Peduncle width at the first spikelet	T27	Cm
Rachis length (from petiole to terminal leaflet base)	T5	cm	Ramified bunch's part length	T28	Cm
Rachis length (spiny part)	T6	cm	Spikelet's length at the bunch's bottom	T29	Cm
Rachis length (1st Leaflet to terminal leaflet base)	T7	cm	Spikelet's length at the bunch's middle	T30	Cm
Rachis thickness	T8	cm	Spikelet's length at the bunch's top	T31	Cm
petiole length	T9	cm	number of fruits per cluster (spikelet)	T32	G
Petiole width	T10	cm	weight of fruits per cluster (spikelet)	T33	G
Number of pinnae (right side)	T11	count	number of cluster (spikelet) per bunch	T34	Count
Number of pinnae (left side)	T12	count	weight of date fruits per bunch	T35	Kg
Basal pinnae length	T13	cm	average weight of date fruits per tree	T36	Kg
Basal pinnae width	T14	cm	No. Bunch per tree	T37	Count
Median pinnae length	T15	cm	Fruit weight	T38	G
Median pinnae width	T16	cm	Fruit length	T39	Mm
Apical pinnae length	T17	cm	Fruit width	T40	Mm
Apical pinnae width	T18	cm	Pulp weight	T41	G
Terminal leaflet length	T19	cm	Pulp thickness	T42	Mm
Terminal leaflet width	T20	cm	Seed weight	T43	G
Last terminal leaflet number	T21	count	Seed length	T44	Mm
Angle on both sides of the terminal leaflets	T22	degree	Seed width	T45	Mm
Spine number	T23	count			

DATA ANALYSES

Morphological data analysis

A total of 45 morphological traits' data were used to calculate mean values, analysis of variance (ANOVA) using SPSS software version 28.0.1. The homogeneity of variances (HOV) of date palm cultivars was calculated using Levene's test in SPSS software to confirm these traits had equal variance or not among the cultivars. Also, vegetative traits data and reproductive data were used separately to create dendrogram and Principal component analysis (PCA) biplot loading to discriminate the relationship between date palm cultivars. PCA of biplot loading was performed to detect correlation between morphological traits and date palm cultivars using "ward" method in R package; on PCA biplot loading, small angle

between any two vectors indicates a strong positive correlation between variables and large angle between any two vectors indicates negative association between two variables. Besides, vectors (arrows) that have same direction and longer on PCA quadrant showed strongly correlated variables and vice versa and due to these vectors showed their contribution to discriminate and classify variables on PCA axes (Ginanjar *et al.*, 2017).

RESULTS

PCA analysis and morphological relations of cultivars

In this study, mean values of all morphological traits showed variations among date palm cultivars (Table 2).

Table 2. Mean value of 45 morphological traits of 11 date palm cultivars.

Date palm cultivars												
Traits	Berhee	Khalas	Medjool England	Medjool Israel	Shishi	Zamlli	Ashal Hassa	Al Hassa	Khyara	Saggii	Kharda wy	Jarvis - male
T1	207.3 ± 10.5	165.5 ± 24.7	165.5 ± 24.7	160.3 ± 27.2	256.0 ± 52.2	230.5 ± 74.1	193.0 ± 20.3	160.8 ± 34.3	204.0 ± 16.1	122.3 ± 19.1	142.0 ± 57.0	
T2	238 ± 9.85	232.3 ± 10.0	221.3 ± 37.0	171.3 ± 28.7	256.3 ± 52.2	266.3 ± 25.6	248.5 ± 20.9	202.5 ± 9.6	255.0 ± 23.8	205.0 ± 10.0	230 ± 35.6	
T3	463.5 ± 8.4	424.8 ± 31.6	349.0 ± 32.1	326.5 ± 31.1	360.6 ± 16.7	421.5 ± 69.0	398.3 ± 55.1	479.8 ± 9.4	419.0 ± 22.4	361.3 ± 26.3	353.0 ± 109.3	
T4	89.3 ± 4.3	91.3 ± 3.8	103.5 ± 11.7	101.5 ± 8.2	97.5 ± 4.2	86.0 ± 4.9	86.5 ± 5.8	82.8 ± 3.6	116.6 ± 9.1	77.3 ± 2.9	98.5 ± 11.7	
T5	435.5 ± 10.6	411.0 ± 31.2	331.8 ± 28.5	306.3 ± 31.0	342.3 ± 18.6	379.8 ± 72.9	364.0 ± 26.5	453.3 ± 11.6	391.5 ± 25.4	340.8 ± 27.5	321.0 ± 116.2	
T6	104.5 ± 10.8	113.3 ± 13.4	84.0 ± 16.6	88.3 ± 18.8	91.8 ± 13.6	83.0 ± 132.8	97.0 ± 8.7	98.0 ± 11.9	76.0 ± 13.6	90.0 ± 10.2	73.5 ± 31.5	
T7	293.3 ± 14.0	267.5 ± 22.8	250.8 ± 53.1	208.5 ± 19.1	232.3 ± 9.7	277.8 ± 40.0	248.3 ± 16.7	314.5 ± 7.9	290.5 ± 23.4	221.0 ± 22.7	233.5 ± 86.2	
T8	10.1 ± 1.0	9.0 ± 0.4	8.5 ± 0.6	8.5 ± 1.0	10.0 ± 0.0	9.5 ± 0.6	10.0 ± 0.0	8.5 ± 0.6	10.0 ± 0.0	6.3 ± 0.5	9.0 ± 1.2	
T9	170.3 ± 8.5	157.0 ± 14.9	123.3 ± 14.2	118.0 ± 16.0	128.5 ± 13.5	143.8 ± 30.1	150.0 ± 40.1	165.3 ± 12.7	128.5 ± 19.7	139.5 ± 11.5	119.5 ± 23.4	
T10	13.1 ± 1.7	11.5 ± 1.1	11.3 ± 2.3	9.8 ± 1.3	14.1 ± 0.8	13.3 ± 1.7	14.3 ± 0.8	13.0 ± 2.3	14.0 ± 1.1	10.0 ± 0.5	11.0 ± 1.5	
T11	100.5 ± 5.4	96.0 ± 2.1	84.0 ± 4.5	81.8 ± 7.5	89.5 ± 3.5	93.8 ± 15.6	94.8 ± 5.1	109.8 ± 5.3	106.3 ± 3.9	90.0 ± 1.4	68.5 ± 25.6	
T12	103.3 ± 5.9	96.5 ± 1.0	86.0 ± 2.2	84.3 ± 6.4	89.5 ± 4.9	93.3 ± 15.6	93.5 ± 3.1	110.8 ± 5.1	107.5 ± 3.7	92.0 ± 1.3	69.0 ± 25.2	
T13	51.3 ± 2.6	52.8 ± 1.7	59.8 ± 6.1	61.0 ± 11.5	48.6 ± 3.9	46.0 ± 4.8	42.3 ± 1.5	48.0 ± 3.5	67.3 ± 2.0	36.8 ± 1.3	52.3 ± 12.4	
T14	2.1 ± 0.4	2.8 ± 0.6	2.1 ± 0.6	2.1 ± 0.4	2.1 ± 0.3	2.3 ± 0.6	2.0 ± 0.3	0.8 ± 0.2	1.2 ± 0.2	1.4 ± 0.2	2.0 ± 0.3	
T15	47.5 ± 1.3	44.5 ± 1.3	50.8 ± 3.3	49.5 ± 0.6	46.0 ± 3.0	47.0 ± 1.4	40.5 ± 1.7	38.5 ± 2.6	56.3 ± 3.3	39.0 ± 0.0	52.0 ± 5.8	
T16	3.4 ± 0.6	3.4 ± 0.4	3.3 ± 0.2	3.4 ± 0.5	3.8 ± 0.4	3.8 ± 0.4	3.4 ± 0.2	2.9 ± 0.2	2.9 ± 0.4	2.6 ± 0.2	3.1 ± 0.2	
T17	40.3 ± 6.1	31.5 ± 3.3	35.3 ± 4.6	37.5 ± 1.7	38.8 ± 5.1	40.8 ± 3.5	38.3 ± 4.2	39.5 ± 3.7	44.5 ± 7.2	30.0 ± 3.8	46.8 ± 8.8	
T18	2.4 ± 0.4	2.8 ± 0.2	2.9 ± 0.1	3.0 ± 0.2	3.1 ± 0.5	3.4 ± 0.3	2.7 ± 0.3	2.4 ± 0.4	2.5 ± 0.3	2.2 ± 0.3	3.0 ± 0.2	
T19	11.0 ± 0.7	14.2 ± 2.5	16.7 ± 5.0	22.6 ± 1.1	23.5 ± 4.1	26.4 ± 0.9	18.9 ± 1.7	25.8 ± 1.3	29.3 ± 2.6	19.5 ± 2.2	32.9 ± 11.8	
T20	1.1 ± 0.3	0.8 ± 0.1	1.2 ± 0.5	1.1 ± 0.5	1.6 ± 0.5	1.3 ± 0.4	1.7 ± 0.5	1.0 ± 0.0	1.1 ± 0.0	1.4 ± 0.4	1.8 ± 0.6	
T21	2.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.0	3.0 ± 0.3	2.5 ± 0.6	3.0 ± 0.0	3.0 ± 0.0	2.75 ± 0.5	3.0 ± 0.0	3.0 ± 0.0	
T22	93.3 ± 4.7	81.3 ± 6.3	90.8 ± 6.5	89.5 ± 4.9	83.0 ± 4.4	93.3 ± 4.3	86.0 ± 4.5	91.5 ± 2.4	89.8 ± 2.4	84.8 ± 3.5	82.8 ± 2.1	
T23	30.8 ± 1.7	28.8 ± 1.3	29.3 ± 4.6	30.5 ± 2.4	21.0 ± 0.8	24.8 ± 4.2	34.3 ± 1.1	19.8 ± 3.3	24.3 ± 2.8	19.8 ± 0.5	27.0 ± 6.0	
T24	0.6 ± 0.2	0.8 ± 0.1	1.0 ± 0.2	0.8 ± 0.1	1.0 ± 0.2	1.0 ± 0.2	0.9 ± 0.1	0.4 ± 0.2	0.3 ± 0.0	0.3 ± 0.1	1.0 ± 0.1	
T25	13.4 ± 1.9	18.2 ± 2.6	11.6 ± 3.1	10.8 ± 1.0	13.3 ± 2.7	10.8 ± 0.7	12.7 ± 1.7	13.5 ± 3.6	7.4 ± 0.6	8.9 ± 1.2	15.6 ± 4.9	
T26	186.0 ± 9.7	137.0 ± 4.2	65.0 ± 6.4	62.0 ± 28.4	116.3 ± 15.5	132.0 ± 17.8	105.5 ± 19.2	126.0 ± 25.8	131.0 ± 10.1	114.0 ± 7.6	0	
T27	8.4 ± 1.6	7.5 ± 0.7	8.6 ± 1.1	8.6 ± 1.8	8.3 ± 1.3	8.5 ± 1.3	9.0 ± 1.4	8.5 ± 2.1	9.3 ± 1.3	6.2 ± 0.5	0	
T28	32.0 ± 6.4	17.5 ± 3.7	24.3 ± 6.5	32.8 ± 14.0	22.0 ± 11.1	19.5 ± 4.2	14.5 ± 11.2	30.5 ± 11.1	16.8 ± 1.7	27.3 ± 8.2	0	
T29	54.5 ± 9.8	52.8 ± 1.9	49.0 ± 9.0	49.8 ± 13.9	53.0 ± 5.9	46.5 ± 6.8	48.3 ± 11.3	48.5 ± 13.4	52.0 ± 2.4	38.0 ± 1.8	0	

T30	49.3 ± 5.9	45.1 ± 3.6	40.3 ± 8.4	35.0 ± 11.9	50.0 ± 6.5	44.8 ± 8.3	43.8 ± 9.9	39.0 ± 10.0	49.5 ± 4.7	30.5 ± 2.6	0
T31	26.3 ± 3.1	38.3 ± 4.6	32.3 ± 8.3	25.5 ± 7.3	37.8 ± 12.3	29.0 ± 10.9	40.0 ± 9.6	28.5 ± 6.6	40.0 ± 7.1	23.8 ± 2.1	0
T32	11.1 ± 2.5	10.8 ± 3.8	10.2 ± 1.3	7.6 ± 0.6	16.8 ± 7.1	9.2 ± 2.1	15.7 ± 5.0	13.3 ± 4.1	9.1 ± 0.7	15.9 ± 6.8	0
T33	174.0 ± 117.5	210.1 ± 122.8	198.3 ± 115.4	170.5 ± 25.2	1188.5 ± 77.5	200.7 ± 22.0	203.5 ± 104.3	338.3 ± 123.2	176.0 ± 27.2	209.6 ± 108.1	0
T34	66.3 ± 5.9	52.3 ± 9.3	61.3 ± 5.3	68.5 ± 10.5	68.5 ± 9.3	53.3 ± 3.3	66.8 ± 36.3	65.5 ± 15.2	57.3 ± 11.9	52.3 ± 6.4	0
T35	8.4 ± 4.1	3.8 ± 0.6	3.2 ± 1.3	5.5 ± 3.1	3.4 ± 1.4	4.0 ± 0.6	5.8 ± 3.0	6.7 ± 3.6	4.2 ± 1.8	3.4 ± 2.1	0
T36	131.5 ± 89.94	17.5 ± 7.67	22.75 ± 6.46	31.05 ± 25.33	11.75 ± 10.56	34.25 ± 19.36	38.75 ± 26.95	31.5 ± 16.46	38.75 ± 17.73	6.75 ± 3.72	0
T37	17.0 ± 9.1	4.3 ± 3.9	7.8 ± 2.3	4.8 ± 3.5	3.5 ± 3.0	6.8 ± 5.2	5.8 ± 2.5	5.0 ± 0.8	7.0 ± 4.1	2.5 ± 1.9	0
T38	3.6 ± 0.1	3.7 ± 0.7	5.0 ± 2.6	2.2 ± 0.6	2.6 ± 1.1	4.9 ± 1.6	6.2 ± 2.3	7.6 ± 2.2	2.2 ± 0.9	8.6 ± 1.5	0
T39	2.2 ± 0.1	2.2 ± 0.1	3.2 ± 1.2	2.5 ± 0.4	2.5 ± 0.5	3.2 ± 0.3	2.9 ± 0.7	3.7 ± 0.2	3.2 ± 0.4	4.2 ± 0.4	0
T40	1.3 ± 0.03	1.3 ± 0.06	1.3 ± 0.4	1.0 ± 0.3	1.0 ± 0.2	1.6 ± 0.3	1.6 ± 0.2	2.1 ± 0.1	1.4 ± 0.2	2.1 ± 0.5	0
T41	2.9 ± 0.2	3.1 ± 0.7	4.2 ± 2.3	1.5 ± 0.7	1.8 ± 1.1	2.2 ± 1.2	5.5 ± 2.0	6.2 ± 2.7	0.9 ± 0.8	6.6 ± 1.2	0
T42	1.2 ± 0.0	0.2 ± 0.0	0.5 ± 0.2	0.5 ± 0.1	0.5 ± 0.1	0.5 ± 0.1	0.3 ± 0.2	0.4 ± 0.1	0.1 ± 0.0	0.4 ± 0.2	0
T43	0.6 ± 0.0	0.7 ± 0.1	0.8 ± 0.2	0.7 ± 0.1	0.9 ± 0.1	1.1 ± 0.2	0.7 ± 0.1	1.0 ± 0.1	0.9 ± 0.3	0.8 ± 0.1	0
T44	1.3 ± 0.0	1.3 ± 0.1	1.8 ± 0.5	1.6 ± 0.2	1.8 ± 0.4	2.0 ± 0.2	1.5 ± 0.2	2.1 ± 0.2	2.1 ± 0.2	1.7 ± 0.5	0
T45	0.5 ± 0.02	0.5 ± 0.02	0.5 ± 0.11	0.4 ± 0.03	0.4 ± 0.04	0.8 ± 0.12	0.6 ± 0.2	0.7 ± 0.14	0.7 ± 0.06	0.8 ± 0.20	0

PCA analysis on the first component accounted 37% in combined vegetative and reproductive traits and the second and third components accounted 29% and 32% of variations in the morphological traits of the cultivars, respectively (Table 3). Additionally, PCA biplot analysis revealed that correlation of morphological traits and location of cultivars on PCA plane (Fig. 1).

In this study, strongly correlated morphological traits and cultivars had been laid together in the same space and uncorrelated traits and cultivars were located in different space on PCA plane (Fig. 1a, b, c). Consequently, PCA biplot loading concept on the above overall morphological traits data analysis showed strongly correlated morphological traits and cultivars were placed on the same PCA plane as follow: rachis length (spiny part), ramified bunch's part length, number of fruits per cluster (spikelet), weight of fruits per cluster (spikelet), number of cluster (spikelet) per bunch, fruit weight, fruit length, pulp weight, pulp thickness, seed weight, seed length and seed width are positively correlated with Khyara and

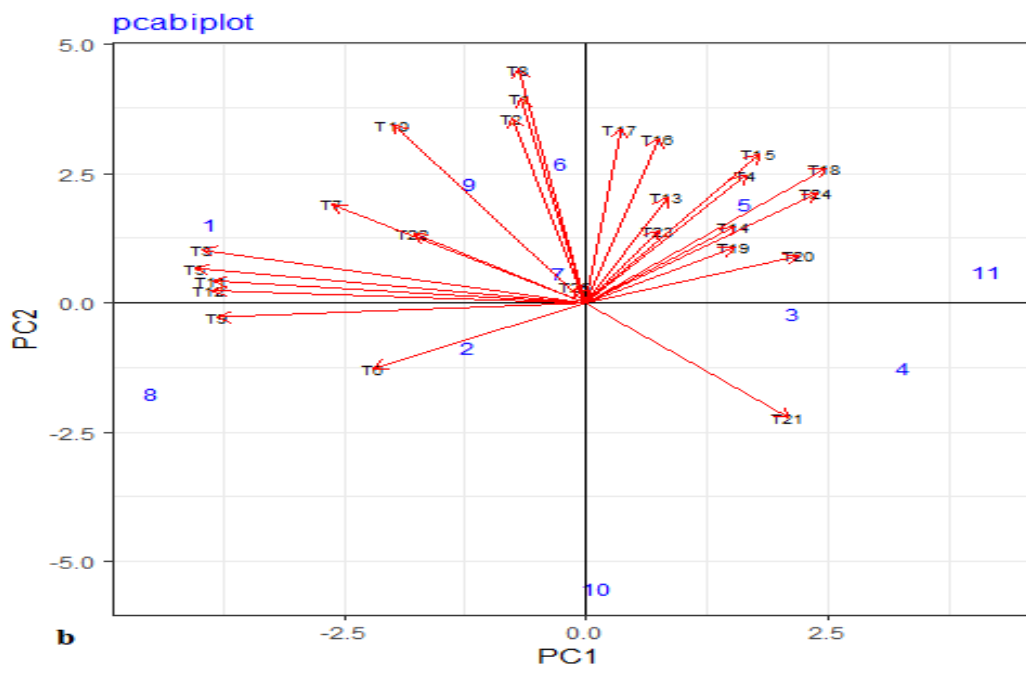
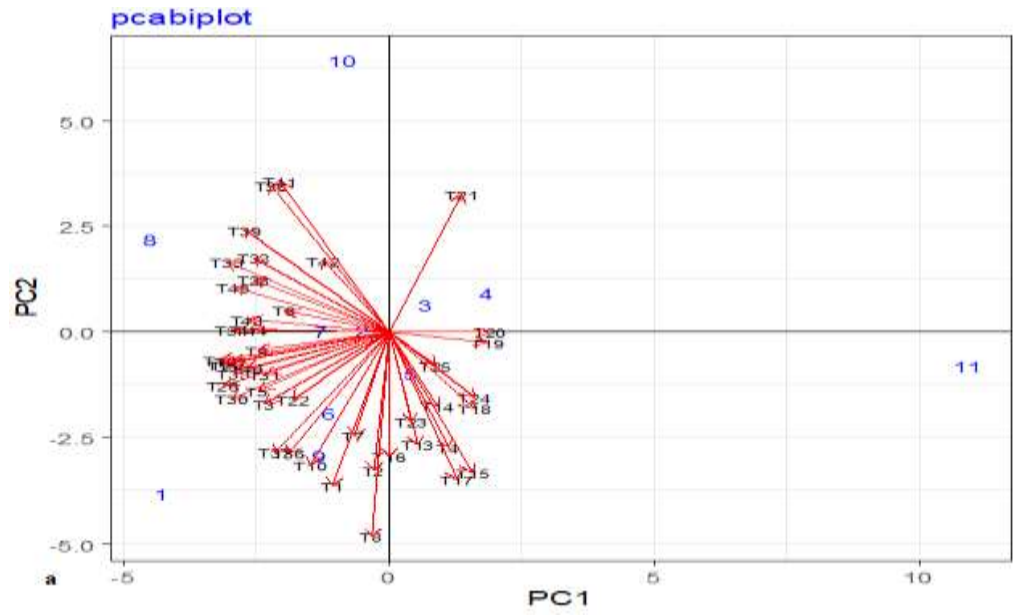
Khadrawy date palm cultivars; rachis length (spiny part), number of cluster (spikelet) per bunch, seed weight and seed length with Khalas and Ashal Al Hassa; frond width at the middle, basal pinnae length, basal pinnae width, median pinnae length, apical pinnae length, apical pinnae width, terminal leaflet length, terminal leaflet width, middle spine width and middle spine length are correlated with Shishi and Jarvis; trunk height, trunk circumference, frond length, rachis length (from petiole to terminal leaflet base), rachis thickness, petiole width, number of pinnae (left side), angle on both sides of the terminal leaflets, peduncle length from its base to the first spikelet, peduncle width at the first spikelet, spikelet's length at the bunch's bottom, spikelet's length at the bunch's middle, spikelet's length at the bunch's top, weight of date fruits per bunch, average weight of date fruits per tree, number of bunch per tree and fruit width are with Barhee, Zamlli and Saggii (Fig. 1a).

Table 3. Seven principal components of date palm cultivars using morphological traits.

	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Overall traits							
Standard deviation	4.10	2.75	2.40	2.21	1.90	1.51	1.31
Proportion of variance	0.37	0.17	0.12	0.11	0.08	0.05	0.04
Cumulative proportion	0.37	0.54	0.67	0.78	0.85	0.91	0.94
Vegetative traits							
Standard deviation	2.71	2.35	2.06	1.61	1.28	1.13	1.02
Proportion of variance	0.29	0.22	0.17	0.10	0.06	0.05	0.04
Cumulative proportion	0.29	0.51	0.68	0.79	0.86	0.91	0.95
Reproductive traits							
Standard deviation	2.52	2.07	1.73	1.50	1.32	0.94	0.87
Proportion of variance	0.32	0.22	0.15	0.12	0.09	0.45	0.04
Cumulative proportion	0.32	0.53	0.68	0.80	0.91	0.92	0.96

In vegetative trait data analysis PCA biplot also represented cultivars clustering with similar patterns of traits on PCA quadrant like trunk height, trunk circumference, frond length, rachis length (from petiole to terminal leaflet base), rachis length (1st leaflet to terminal leaflet base, rachis thickness, petiole width, number of pinnae (right side), number of pinnae (left side), angle on both sides of the terminal leaflets and middle spine length are positively correlated with Barhee, Zamlli, Ashal Al Hassa, Saggii; frond width at the middle, basal pinnae length, basal pinnae width,

median pinnae length, median pinnae width, apical pinnae length, apical pinnae width, terminal leaflet length, terminal leaflet width, spine number with Shishi and Jarvis; Rachis length (spiny part) and petiole length with Khalas and Khyara (Fig. 1b). In addition, reproductive traits data in PCA analysis have shown strong correlation between peduncle length from its base to the first spikelet, length of the bunch's part length, spikelet's length at the bunch's bottom, number of cluster (spikelet) per bunch, weight of date fruits per bunch, average weight of date fruits per tree, number of bunch per tree, fruit width traits and Barhee; peduncle width at the first spikelet, spikelet's length at the bunch's bottom, spikelet's length at the bunch's middle, spikelet's length at the bunch's top traits and Khalas, Medjool England, Shishi, Saggii; pulp thickness, seed weight, seed length traits and Medjool Israel and Zamlli; number of fruits per cluster (spikelet), weight of fruits per cluster (spikelet), fruit weight, fruit length, pulp weight, seed width traits and Khadrawy (Fig. 1c). However, Medjool England, Medjool Israel, Khadrawy and Jarvis cultivars are distantly positioned from the rest cultivars on PCA plane due to variations in almost all vegetative morphological traits (Fig. 1c) and this also happened similarly in phenogram hierarchical clustering result (Fig. 2b). In addition, last terminal leaflet number trait was the most significant trait to discriminate Medjool Israel and Medjool England from the rest cultivars in PCA analysis of morphological data (Fig. 1a-c).



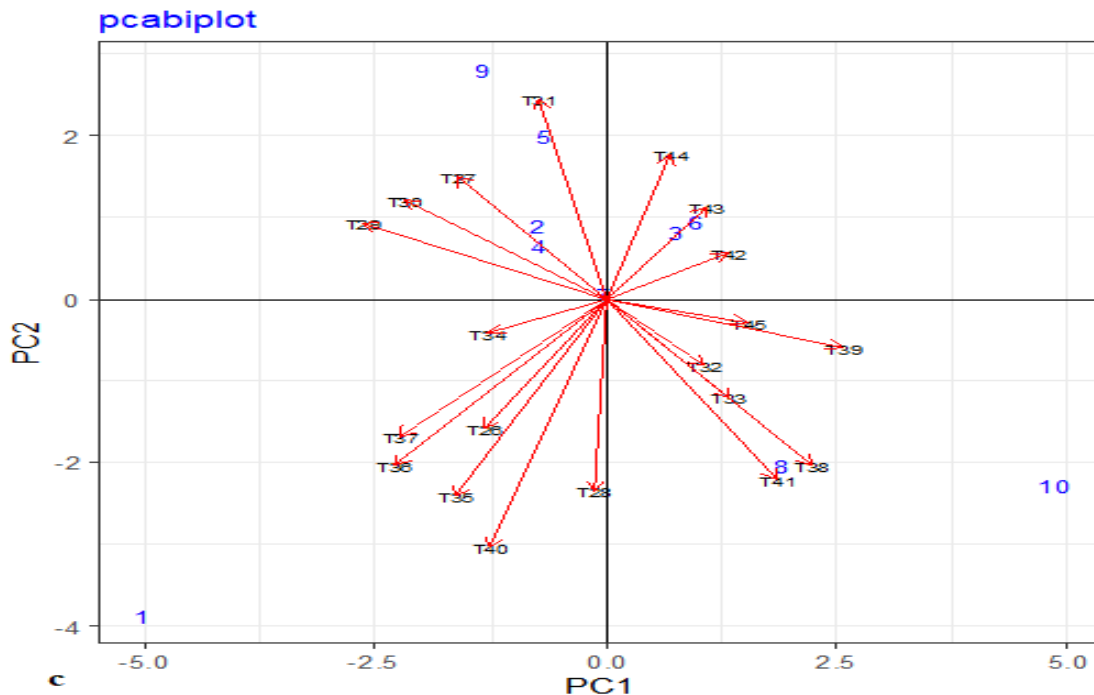
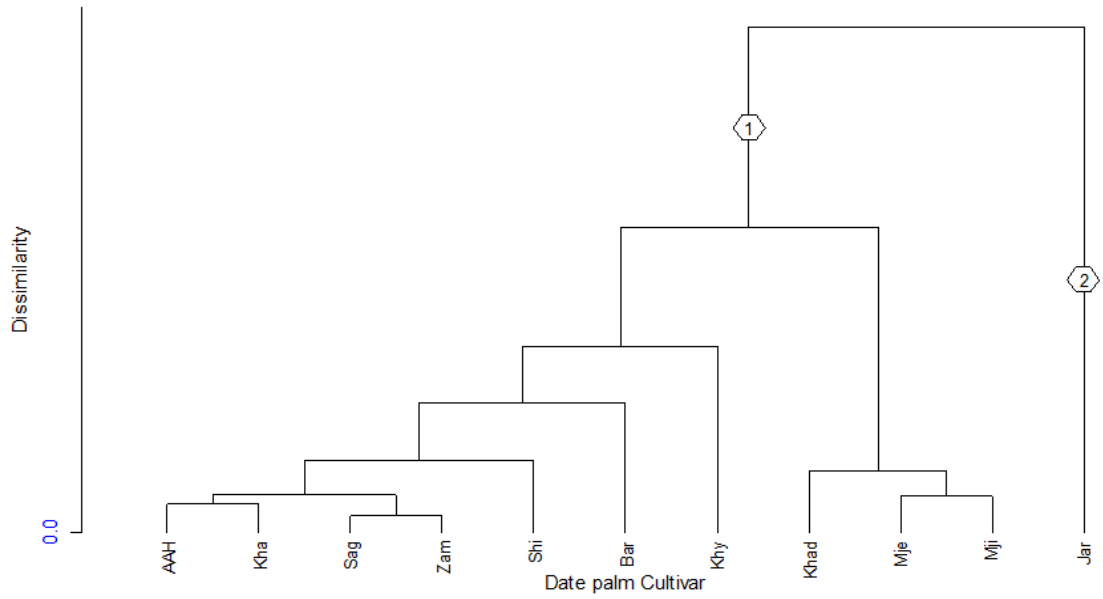


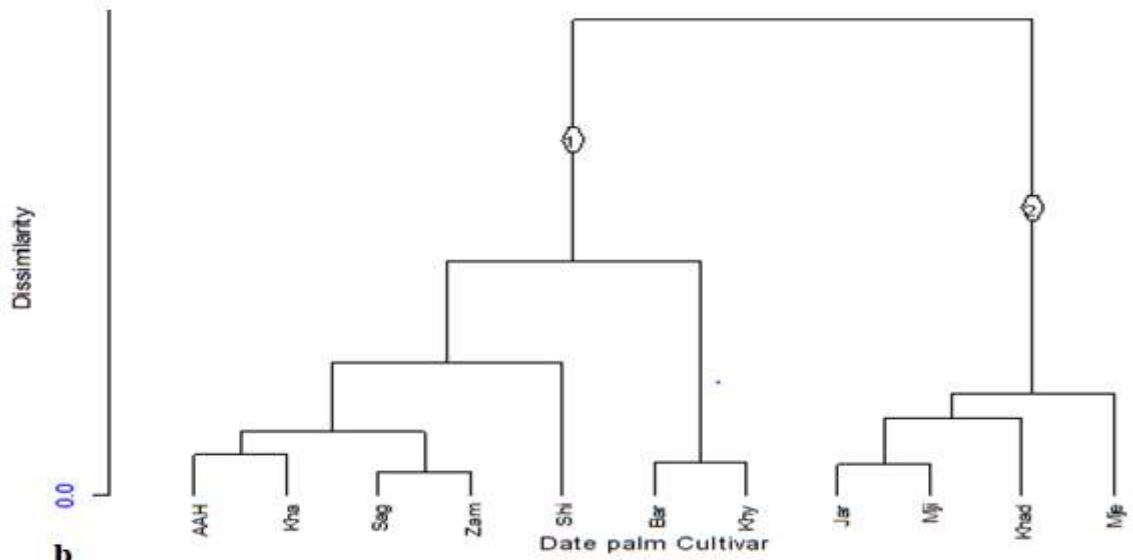
Figure 1. PCA biplots showing the combination of correlated morphological traits and date palm cultivars that are located on the same quadrant according to the cosine angle between any two vectors and direction of vectors a) PCA analysis based on the overall morphological dat; b) PCA analysis based on vegetative dat; c) PCA analysis based on reproductive data. The numbers on PC plane represent date palm cultivars: 1 - Barhee; 2 - Khalas; 3 - Medjool Israel; 4 - Medjool England; 5 - Shishi; 6 - Zamlli; 7 - Ashal Al Hassa; 8 - Khyara; 9 - Saggii; 10 - Khadrawy; 11 - Jarvis (male) and traits are represented by arrows or vectors

In hierarchical cluster analysis based on overall morphological traits, date palm cultivars were divided mainly into male (Jarvis cultivar) and females (Fig. 2a). Females were classified into two groups; one group contained Khadrawy, Medjool England and Medjool Israel and the second group was also determined by two sub-clusters. The first sub-cluster included Ashal Al Hassa, Khalas, Saggi, Zamlli, Shishi and Barhee and the second sub-cluster contained only Khyara. The phenogram (Fig. 1a) revealed that there was no genetic dissimilarity between Khalas and Ashal Al Hassa; Zamlli and Saggii; Medjool Israel and Medjool England, while the rest cultivars exhibited genetic variation. Clustering of cultivars based on vegetative traits showed two main categories. The first one also further divided in to two sub-categories: Ashal Al Hassa, Khalas, Saggi, Zamlli and Shishi were clustered together and Barhee and Khyara represented another sub-category of the

first sub-cluster of a phenogram (Fig. 2b). The phenogram in Fig. 2b indicated that there was no variability in their vegetative traits between Barhee and Khyara; Khalas and Ashal Al Hassa; Zamlli and Saggii; Medjool Israel and Jarvis whereas Shishi, Khadrawy and Medjool England showed genetic dissimilarity and shared some vegetative traits with others. In cluster analysis based on reproductive traits two clusters were observed. The first cluster was further divided into two sub-clusters: one sub-cluster contained only Khyara and the second contained Medjool Israel, Medjool England, Khadrawy, Saggii, Zamlli, Khalas, Shishi and Asha Al Hassa while the second cluster comprises only Barhee cultivar (Fig. 2c). According to the phenogram in Fig. 2c, Khalas and Zamlii; Shishi and Ashal Al Hassa; Medjool Israel and Medjool England showed high similarity and they shared most of their reproductive traits.



a



b

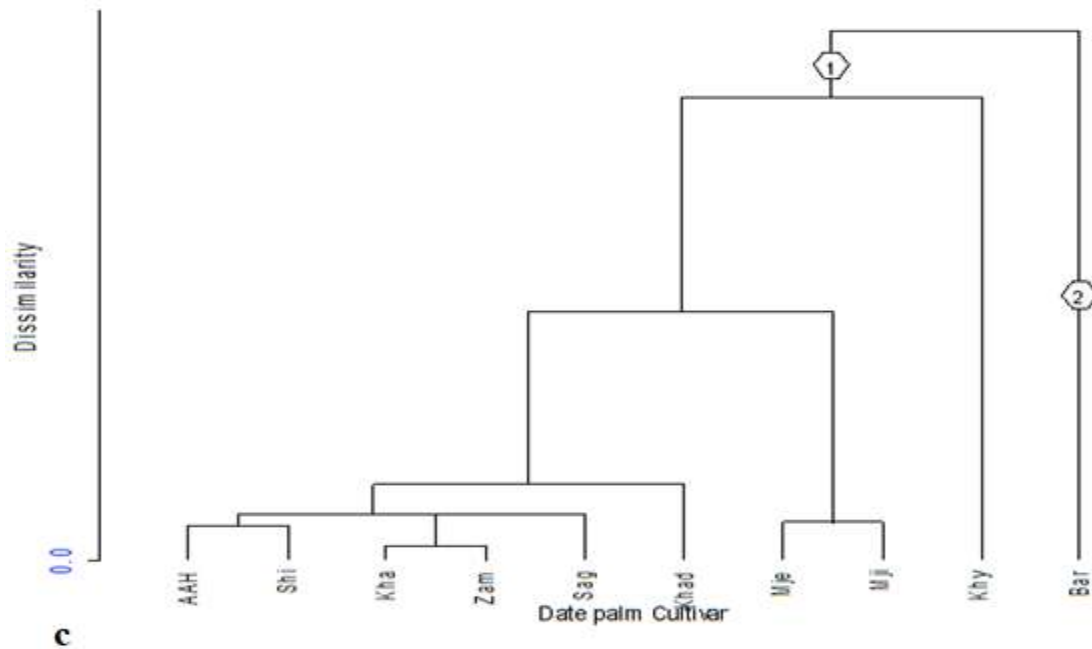


Figure 2. Hierarchical clustering indicating the relationships among date palm cultivars a) based on the overall morphological traits data; b) based on vegetative traits data; and c) based on reproductive traits data that were used in this study. The codes on the tree represent cultivars: AAH - Ashal Al Hassa; Kha - Khalas; Sag - Saggi; Zam - Zamilli; Shi - Shishi; Bar - Barhee; Khy - Khyara; Jar - Jarvis (male); Mji - Medjool Israel; Mje - Medjool England; Khad - Khadrawy.

ANOVA and homogeneity of variances in date palm cultivars

Analysis of variance (ANOVA) detected significant differences among cultivars at $p < 0.05$ or $p < 0.001$ in all morphological traits except traits of rachis length of spiny part and fruit width (Table 4). According to Levene's statistics result (Table 5) 12 traits as mentioned here: frond width at the middle, petiole length, petiole width, basal pinnae width, median pinnae width, apical pinnae

length, apical pinnae width, angle on both sides of the terminal leaflets, middle spine width, peduncle width at the first spikelet, ramified bunch's part length and spikelet's length that the bunch's top showed insignificant differences among date palm cultivars whereas, the rest of 23 traits had $p < 0.05$ value, so we had sufficient evidence to say that the variance of these traits among cultivars is significantly different.

Table 4. ANOVA result among 11 date palm cultivars using morphological traits data

Traits	Sum of Squares	Mean Square	F	Sig.	Traits	Sum of Squares	Mean Square	F	Sig.
Trunk height	63924.1	6392.4	4.6	0	Middle spine width	2.94	0.29	13.7	0
Trunk circumference	30602.6	3060.2	4.9	0	Middle spine length	358.62	35.7	5.58	0
Fronde length	99677.7	9967.7	4.5	0	Peduncle length from its base to the first spikelet	96665.7	9666.6	39.24	0
Fronde width at the middle	4916.6	491.66	9.8	0	Peduncle width at the first spikelet	276.41	27.6	16.02	0
Rachis length (from petiole to terminal leaflet base)	91467.2	9146.7	4.1	0.001	Ramified bunch's part length	3649.91	365.9	5.356	0
Rachis length (spiny part)	5547.01	554.71	1.7	0.119*	Spikelet's length at the bunch's bottom	9608.23	960.8	13.65	0
Rachis length (1st Leaflet to terminal leaflet base)	44162.4	4416.24	4.3	0.001	Spikelet's length at the bunch's middle	8201.65	820.7	15.08	0
Rachis thickness	51.26	5.13	11.7	0	Spikelet's length at the bunch's top	6660.68	666.07	11.96	0
petiole length	13650.1	1365.01	3.2	0.006	number of fruits per cluster	890.64	89.064	5.918	0
Petiole width	107.28	10.73	5.1	0	weight of fruits per cluster	240203	24020.3	3.009	0.008
Number of pinnae (right side)	5330.7	533.07	5.3	0	number of clusters per bunch	15285.7	1528.6	8.062	0
Number of pinnae (left side)	5445.2	544.52	5.8	0	weight of date fruits per bunch	190.2	19.02	3.498	0.003
Basal pinnae length	3011	301.1	8.5	0	average weight of date fruits per tree	49402.8	4940.3	3.876	0.002
Basal pinnae width	12.47	1.25	7.8	0	No. bunch per tree	742.1	74.2	3.5	0.003
Median pinnae length	1256.5	125.65	17.2	0	Fruit weight	259.2	25.92	11.68	0
Median pinnae width	5.86	0.59	4.3	0.001	Fruit length	48.1	4.81	20.48	0
Apical pinnae length	1005.7	100.57	3.7	0.002	Fruit width	43.4	4.34	1.38	0.232*
Apical pinnae width	5.45	0.55	5.4	0	Pulp weight	192.2	19.22	9.21	0
Terminal leaflet length	1743.6	174.36	9.4	0	Pulp thickness	1.3	0.13	10.19	0
Terminal leaflet width	4.23	0.42	2.2	0.043	Seed weight	3.3	0.33	16.26	0
Last terminal leaflet number	4.14	0.41	7.8	0	Seed length	17.1	1.71	10.41	0
Angle on both sides of the terminal leaflets	769.91	76.99	4.1	0.001	Seed width	1.9	0.19	16.96	0
Spine number	945.18	94.52	9.8	0					

*Indicates insignificant value at $p \leq 0.05$

Table 5. Test of homogeneity of variances (HOV) on morphological traits of 11 date palm cultivars of this study.

Morphological traits	Levene Statistic	df1	df2	Sig.	Morphological traits	Levene Statistic	df1	df2	Sig.
Trunk height	2.279	10	33	0.037	Middle spine width	1.438	10	33	0.207*
Trunk circumference	2.138	10	33	0.049	Middle spine length	3.009	10	33	0.008
Fronde length	6.85	10	33	0	Peduncle length from its base to the first spikelet	2.231	10	33	0.041
Fronde width at the middle	1.525	10	33	0.175**	Peduncle width at the first spikelet	1.432	10	33	0.21*
Rachis length (from petiole to terminal leaflet base)	14.257	10	33	0	Ramified bunch's part length	1.79	10	33	0.102*
Rachis length (spiny part)	2.719	10	33	0.015	Spikelet's length at the bunch's bottom	2.969	10	33	0.009
Rachis length (1st leaflet to terminal leaflet base)	2.604	10	33	0.019	Spikelet's length at the bunch's middle	1.851	10	33	0.09
Rachis thickness	5.823	10	33	0	Spikelet's length at the bunch's top	2.004	10	33	0.065*
petiole length	1.843	10	33	0.091*	number of fruits per cluster (spikelet)	4.059	10	33	0.001
Petiole width	1.479	10	33	0.191*	weight of fruits per cluster (spikelet)	2.322	10	33	0.034
Number of pinnae (right side)	14.041	10	33	0	number of cluster (spikelet) per bunch	6.091	10	33	0
Number of pinnae (left side)	16.314	10	33	0	weight of date fruits per bunch	4.247	10	33	0.001
Basal pinnae length	2.184	10	33	0.045	average weight of date fruits per tree	4.58	10	33	0
Basal pinnae width	1.835	10	33	0.093*	No. Bunch per tree	5.057	10	33	0
Median pinnae length	6.926	10	33	0	Fruit weight	2.239	10	33	0.04
Median pinnae width	1.246	10	33	0.3*	Fruit length	3.554	10	33	0.003
Apical pinnae length	1.124	10	33	0.374*	Fruit width	8.269	10	33	0
Apical pinnae width	1.31	10	33	0.2658*	Pulp weight	2.966	10	33	0.009
Terminal leaflet length	3.801	10	33	0.002	Pulp thickness	3.049	10	33	0.008
Terminal leaflet width	2.976	10	33	0.009	Seed weight	2.533	10	33	0.022
Last terminal leaflet number	22.6	10	33	0	Seed length	4.288	10	33	0.001
Angle on both sides of the terminal leaflets	0.764	10	33	0.662*	Seed width	4.23	10	33	0.001
Spine number	5.966	10	33	0					

*Indicates insignificant value at $p \leq 0.05$

DISCUSSION

For sustainable utilization of plant genetic resources, it is necessary to understand and determine the natural relationships among plant varieties and cultivars based on scientific knowledge and principles. Genetic identification of plant varieties using morphological traits can provide evidences on the bases of their evolutionary relationships. The purpose of this study was to identify and evaluate the relationships of date palm cultivars using morphological markers. In this study, significant

variation was presented among date palm cultivars as revealed by PCA, dendrogram, and ANOVA and HOV analysis results. Additionally, all morphological traits showed heterogeneity among date palm cultivars except rachis length of spiny part and fruit width traits which showed insignificant value at $p \leq 0.05$. Bedjaoui and Benbouza, (2018) suggested that phenotypic variation was exhibited among Algerian date palm cultivars in both vegetative and reproductive characters in which reproductive characters showed stronger dissimilarity among cultivars than vegetative traits. This result is in agreement with the present study. According to ANOVA

result of this study pinnae number, pinnae width and length, and trunk height traits were showed significant values at $p \leq 0.05$ whereas Boudeffeur *et al.*, (2021) reported that these traits showed nonsignificant value at $p \leq 0.05$. Additionally, Khalilia *et al.*, (2022) and Allam *et al.* (2021) also reported the fruits width showed significant variation between date palm cultivars while this trait revealed contrary result in this study.

According to PCA biplot loading result of the present study, strong and weak correlation of morphological traits was observed for grouping of date palm cultivars on PC planes or quadrants. PCA biplot loading displayed that those vegetative traits: trunk height, trunk circumference, frond length, rachis length (from petiole to terminal leaflet base), number of pinnae (right side) and number of pinnae (left side) were useful traits to separate Barhee cultivar from the rest date palm cultivars. This finding is consistent with Eissa *et al.* (2009) who reported that Barhee cultivar differentiated from the rest genotypes mainly by trunk height, trunk circumference, frond length, rachis length (from petiole to terminal leaflet base), number of pinnae. Additionally, in this study the reproductive traits peduncle length from its base to the first spikelet, ramified bunch's part length, number of cluster (spikelet) per bunch, weight of date fruits per bunch, average weight of date fruits per tree, number of bunches per tree and fruit width were the most useful and correlated parameters to discriminate Berhee cultivar from others. On the other hand, Hammadi *et al.* (2009); Elsafy *et al.* (2015); El -Sharabasy and Rizk (2005); Ouarda *et al.* (2012); Abdelkrim *et al.* (2020) have reported fruit, spine, frond and trunk morphological traits of date palm are the most important parameters for discrimination and identification of cultivars. In general, based on the result of this study we observed that there is variation of morphological traits that briefly distinguish between date palm cultivars particularly for those closely related ones. This implicate there exist high genetic variation among date palm population in Ethiopia, this is a rich genetic wealth could be seriously studied to harness them for genetic improvement. In general, this research is the first work for documenting and identifying the relationships among date palm cultivars in Ethiopia using morphological markers. Consequently, this study could be useful to give shade light on the understanding on

morphological relationships of date palm cultivars and also will assist in selecting of cultivars for breeding and future agronomic important traits improvement of this valuable crop in Ethiopia.

CONCLUSIONS

Morphological traits have shown genetic variations among date palm cultivars grown at Melka Werer Agricultural Research Center. The overall statistical and clustering analysis results of this study indicated the phylogenetic relationships of date palm cultivars. Generally, our finding on the morphological trait analysis results will be an important groundwork for documentation, genetic improvement and conservation, and for further agronomic important traits studies of date palm in Ethiopia.

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