

Diversity among *Coffea arabica* populations in southwestern Saudi Arabia as revealed by their morphometric features

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Abstract

Coffee (*Coffea arabica* L.) is one of the most important agricultural commodities traded worldwide. The livelihoods of millions of households in Asia, Africa and America depend on it. The sustainability of the supply chain of this crop is increasingly under threat due to the impact of climate change in the main producing countries in the tropics and sub-tropics. The resilience of these agro-ecosystems will depend on the ability of breeders to develop new coffee varieties that can better adapt to changing environmental conditions. Therefore, studying the diversity of coffee populations in the Arabian Peninsula could reveal agronomically interesting genotypes that can be exploited in breeding programs. The objective of the study was to evaluate the diversity among coffee populations in southwestern Saudi Arabia using quantitative morphological, pomological and agronomic traits. The analysis of variance of the data showed differences among the accessions for most of the measured quantitative traits. The accessions varied in growth habit, canopy shape and cherry, bean and leaf dimensions. Cherry fresh mass ranged from 96.5 to 234.8 g in 100 cherries while 100-bean dry mass varied from 9.3 to 22.5 g. The hierarchical cluster analysis divided the accessions into four main groups. The study revealed considerable variability among the 61 accessions. Based on this investigation, accessions KSA-7R, KSA-8 and KSA-9R from Tallan valley, KSA20, KSA21 and KSA52 from Fayfa, KSA38 from Eddayar district, KSA10 and KSA60 from Assir region and KSA61 from Jebel Shada are recommended for further investigation for their promising agronomic traits.

Keywords: *Coffea arabica*; diversity; Kholani; Mocha; population structure; Shadawi

Introduction

The genus *Coffea* has more than 100 species but only two are commercially grown, Arabica (*C. arabica* L.) and Robusta (*C. canephora* Pierre ex A. Froehner) (Ferreira *et al.*, 2019). Arabica Coffee is one of the most important agricultural commodities in international trade; more than 125 million people in Asia, Africa and Latin America depend on coffee growing and processing for their livelihoods (Osorio, 2002). Unfortunately, the sustainability of coffee cultivation is increasingly under threat due to the impact of climate change in the main producing countries in the tropics and subtropics (Ahmed *et al.*, 2021). The resilience of these agroecosystems and the economy of the communities dependent on them will depend on the ability of breeders to develop new coffee varieties that can better adapt to changing environmental conditions, increasing pest and disease pressure, and evolving consumer preferences (Lacherme *et al.*, 2009). To achieve this, the breeder needs to have access to a wide genetic pool in search of genes of interest. In the case of Arabica coffee, such diversity can only be found in the species' center of origin in the Ethiopian highlands (Anthony *et al.*, 2001) or, as we hypothesize, in the center of the first domestication of coffee in Yemen and southwestern Saudi Arabia. It is well known that the gene pool of *C. arabica*, a tetraploid species, is narrow (Anthony *et al.*, 2001); therefore, wild diploid coffee species and underutilized ancient tetraploid cultivars could serve as a critical source of genes for coffee breeding programs (Lacherme *et al.*, 2009; Davis *et al.*, 2019). Recent studies by Montagnon *et al.* (2021, 2022a, 2022b) have shown that there is considerable genetic diversity among coffee populations in Yemen. Therefore, studying the diversity of coffee populations in the Arabian Peninsula could reveal agronomically interesting genotypes. The objective of the present study was to evaluate the diversity among coffee populations from various districts in southwestern Saudi Arabia using quantitative morphological, pomological and agronomic traits.

C. arabica has been cultivated for centuries in the southwestern corner of the Arabian Peninsula on mountain terraces between 1000 and 2200 m above sea level (a.s.l.) (Lacherme *et al.*, 2009; Tounekti *et al.*, 2017; Montagnon *et al.*, 2021). In Saudi Arabia, despite the limited acreage dedicated to coffee, the crop is important for the local mountain communities as both a cash crop and a cultural symbol in which they take great pride (Tounekti *et al.*, 2017; Kathurima *et al.*, 2022). Local production accounts for less than 1% of the country's consumption, yet most local coffees are of excellent quality and are in high demand all over the Gulf Region (Gennari *et al.*, 2015; Kathurima *et al.*, 2022). It is well known among coffee connoisseurs that the Arabian Peninsula is the source of the exotic dry processed (natural) coffees commonly known as Mocha or Moka, which have distinctive cup qualities. They are usually full-bodied with winy acidity and deep earthy notes with overtones of dry fruit and spices and a chocolaty finish (Wintgens, 2012; Kathurima *et al.*, 2022). These coffees can potentially fetch high prices on the specialty coffee niche market if cultural practices and post-harvest processes were improved and the cultivars were well characterized and publicized (Kathurima *et al.*, 2022). Unfortunately, these exotic genetic resources are under serious threat to be lost due to periodic droughts and the destruction of the terraces abandoned by their owners (Tounekti *et al.*, 2017), thus the urgent need to characterize these genotypes and conserve them (Montagnon *et al.*, 2022b). Tounekti *et al.* (2019) carried out the first survey of the morphological diversity of Arabica coffee in Saudi Arabia using quantitative traits. They reported a large variability among the populations for most traits even though they studied only 19 accessions.

To ensure stakeholders' support, better financial returns for growers and processors must result from any efforts to preserve these centuries-old genetic resources, and this cannot be done without a sophisticated marketing plan that increases the crop's worth. The standardization of the product, which must adhere to stringent quality standards, is necessary to access the global commodity distribution and marketing networks (Wintgens, 2012; Kathurima *et al.*, 2022). If nurseries continue to raise planting material from seeds of unknown genotypes, which is often the case in Saudi Arabia, it would be hard to meet these market quality standards. Therefore, the planting material offered by the nurseries must be of known cultivars. To this aim,

we conducted the current study to (i) assess the diversity among the Arabica coffee populations in southwestern Saudi Arabia region based on morphometric features of the trees, their leaves and their fruit using multivariate analysis methods, (ii) identify easy to measure features that can be used to identify the cultivars and (iii) identify agronomically interesting genotypes.

Materials and Methods

Collection of plant material

A survey of coffee gardens was carried out in the three administrative regions of Jazan, Assir and Al-Baha in Saudi Arabia (Figure 1). The methodology used for the survey was previously described by Tounekti *et al.* (2017). The survey covered a strip of terraced mountains located between latitudes 17° and 20° N and longitudes 42° and 43° E, covering, from the South to the North, Jebel Fayfa (Fayfa district), Eddayer district, Maadi (Haroub district), Jebel Al-Gahr (Al-Rayth district), Reada valley (Assouda district in Assir region), Mahayel Assir district, Al-Majarda district and Jebel Shada (Al-Mekhwah district of Al-Baha region) (Table 1).

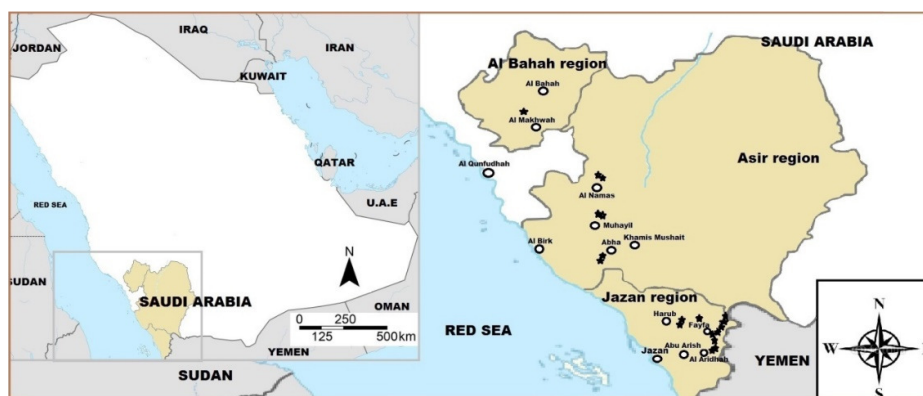


Figure 1. A map of southwestern Saudi Arabia showing the sites where coffee plant material for the study was collected. The collection areas are marked with black stars (adapted with permission from Tounekti *et al.* (2017))

Table 1. Collection sites of coffee germplasm. The sites are located between longitudes 42°22' and 43°07'E

Location/community	District	Region	Altitude (m a.s.l)	Latitude
Khacher/Al-Zoughli	Addayer	Jazan	1,254	17° 18'03"N
Khacher/Al-Guatil	Addayer	Jazan	1,484	17° 19'01"N
Al-Gahr	Al-Rayth	Jazan	1,750; 1,900	17° 38'08"N
JMRDA farm*	Fayfa	Jazan	1,260	17° 15'20"N
JCGR*	Fayfa	Jazan	850	17° 17'13"N
Jebel Fayfa	Fayfa	Jazan	1,440-1,650	17° 15'21"N
Wadi Dafa	Addayer	Jazan	1,254	17° 25'41"N
Jebel Tallan	Addayer	Jazan	1,546-1,672	17° 23'11"N
Sahalil	Haroub	Jazan	950	17° 29'29"N
Maadi	Haroub	Jazan	1,230-1,344	17° 26'37"N
Jebel Hada	Mahayel Assir	Assir	1,503	18° 38'44"N
Reada	Assouda	Assir	1,480-1,594	18° 11'37"N
Wadi Alghil	Al-Majarda	Assir	1,320-1,450	19° 09'35"N
Shada Al-Aala	Al-Mekhwah	Al-Baha	1,548	19° 50'54"N
Shada Al-Asfal	Al-Mekhwah	Al-Baha	1,350	19° 40'30"N

*JMRDA = Jazan Mountain Region Development Authority; JCGR = Jazan Coffee Germplasm Repository

We tagged and sampled 3-4 visibly similar trees to represent each genotype. To minimize the effect of uncontrolled environmental factors, we avoided trees in dense thickets, in proximity of other crops and those with symptoms of water stress, nutritional deficiencies or pest infestations. Each accession was given a code starting with the acronym “KSA” (e.g., KSA-1), but, for the sake of simplicity, we dropped the acronym in the figures. The letter “R” was added to the code of accessions KSA-1-KSA-19, and KSA-45 to indicate that they were sourced from a small, local coffee germplasm collection (Jazan Coffee Germplasm Repository, JCGR) established in the experimental farm of the Jazan Mountain Region Development Authority (JMRDA) in Fayfa district. Most samples were collected during the harvest season from 1 November 2020 to 30 January 2021. In total, we collected cherries and leaves from representative trees of 61 accessions.

Geography of the surveyed area

The climate of the surveyed area falls within the arid (Al-Baha) and semi-arid (Jebel Fayfa) agro-climatic zones characterized by mild winters and warm summers (Eulefeld, 1979; De Pauw, 2002). Climatic data from the JMRDA Meteorological Station indicate that the highest monthly maximum and the lowest monthly minimum temperatures were recorded in June (33 °C) and January (14 °C), respectively (data not shown). The mean maximal monthly rainfall (220 mm) during the same period was recorded in April and the mean minimum in November. Most of the rain is between April and September. Most coffee gardens are located on the more humid west-facing slopes which receive more rain and fog than the eastern slopes. Still rainfall is too limited and erratic to allow for rain-fed coffee cultivation (Sayed *et al.*, 2019). Most coffee gardens receive some supplemental irrigation during the dry period from May to July. The soils were loamy or sandy loam in Jazan and Assir terraces and sandier in Al-Baha sites. They were slightly alkaline with pH ranging from 7.5 to 8.0.

Morphometric measurements

The eighteen morphometric parameters used to describe the trees were presented in Table 2. They were selected based on the International Plant Genetic Resources Institute (IPGRI) (IPDRI, 1996) recommendations and previous similar studies (Anthony *et al.*, 2001; Eskes and Mukred, 1990). For each accession, we collected about 500 g of ripe cherries from each of 3-4 trees. A 100-cherry subsample from each accession was used to measure fruit fresh mass (FFW). A 40-cherry subsample from each accession was used to measure fruit longitudinal diameter or length (FL), fruit equatorial diameter or width (FW), fruit thickness (FT), fruit FL/FW ratio, and Fruit sphericity (FSph) calculated according to Anthony *et al.* (2001) from the equation:

$$FSph = \left(\sqrt[3]{(L \times W \times T)} \right) \div L$$

The reminders of cherry samples were sun-dried for three weeks according to the traditional method of drying coffee by local growers (the natural method). Once dry to a moisture content of 12%, the cherries were hulled with an electric bench-top cherry huller (Model 673 S200, Coffee Laboratory, VA, USA). A 100-bean subsample from each accession was used to determine bean dry mass (BDW). Bean longitudinal diameter or length (BL), bean equatorial diameter or width (BW), bean thickness (BT), bean BL/BW ratio, and bean sphericity (BSph) were measured on a 40-bean subsample from each accession. A digital Vernier calliper with 0.01 mm accuracy (Series 500, Mitutoyo, Japan) was used to measure fruit and bean dimensions. Leaf length (LL), leaf width (LW) and LL/LW ratio were measured on a sample of 40 mature, mid-shoot leaves from each accession. The color of the new leaves (flushes) was given an arbitrary score of 1 if it was green, 2 for pale bronze, 3 for bronze, or 4 for dark bronze or brown (IPDRI, 1996).

For each tree, we also measured the spread (in °) of the crotch angles made by the 9-11th laterals from the top with the tree’s main stem. The lengths of the 2nd- 6th internodes were measured on four primary plagiotropic branches on each tree. The branches were selected at 0.5-1 m tree height, one from each side of the canopy.

The above morphometric measurements were complemented with descriptions of tree height, growth habit, vigour, overall appearance of the canopy, and yield. These latter descriptions were used to help

distinguish the populations but were not included in the statistical analyses because they are not quantitative, or they are confounded by tree age and cultural practices. The scales of the descriptors were adapted from IPIGRI (IPDRI, 1996) and Eskes and Mukred (1990). The trees were classified as dwarf ($H < 1.5$ m), short ($1.5 \leq H < 2.5$ m), medium ($2.5 \leq H \leq 3.5$) or tall ($H > 3.5$). The trees had open, normal or compact canopies. The overall appearance of the canopy was described as conical, intermediate or cylindrical (columnar).

Table 2. Agronomic and morphometric characters used to describe the 61 coffee genotypes considered in the study (adapted from IPDRI (1996) and Eskes and Mukred (1990))

Parameter	Abbreviation
Fruit longitudinal diameter or length (mm)	FL
Fruit equatorial diameter or width (mm)	FW
Fruit thickness (mm)	FT
Fruit FL/FW	FL/FW
Fruit sphericity	FSph
Bean longitudinal diameter (mm)	BL
Bean equatorial diameter (mm)	BW
Bean thickness (mm)	BT
Bean BL/BW	BL/LW
Bean sphericity	BSph
Leaf length (cm)	LL
Leaf width (cm)	LW
Leaf LL/LW	LL/LW
Fresh mass of 100 cherries (g)	FFW
Dry mass of 100 beans (g)	BDW
Color of new flush*	Flsh
Crotch angle: the spread (°) of the crotch angles of the 9-11 th laterals	Angl
Average length (cm) of the internodes of the lower branches	Inod

*Indices were used to evaluate the color of new leaves (flush): 1 for green, 2 for pale bronze, 3 for bronze, 4 for dark bronze or brown.

Statistical analysis

Combining multivariate statistical methods such as principal component analysis (PCA) and hierarchical cluster analysis (HCA) can elucidate the diversity among groups of unknown genotypes based on morphometric or other quantitative characteristics (Johnson and Wichern, 2002). In studies dealing with a fairly large number of independent variables, simple ANOVA methods are inappropriate because interpretation of results becomes hard once the number of factors exceeds three. In such cases, the PCA which is an unsupervised multivariate data analysis method that provides a visual representation of the data on a two-dimensional space is recommended (Stewart *et al.*, 2014). The PCA produces Eigenvectors and factor scores that are used to measure the relative discriminative power of the axes and their associated characters, respectively.

The effects of the treatment (i.e., the genotype) on the measured independent variables were evaluated through a one-factor ANOVA in OriginPro software (OriginLab Corp. MA, USA, 2021), and the means were separated by the Scott-Knott Effect Size Difference test ($P = 0.05$). To assess the relatedness among the 61 accessions, the data were subjected to a multivariate analysis by PCA followed by HCA using OriginPro 2021 software. To bring down the eighteen variables considered in the study to a common scale without distorting the differences in the range of the values, the data were first standardized by subtracting the mean of each variable from the value of the variable for a given accession and dividing by the standard deviation (Stewart *et al.*, 2014). The tables show the original untransformed values. The analysis of variance in the data from morphometric measurements revealed significant differences among the coffee germplasm accessions for all

traits. Therefore, all traits were included in the subsequent multivariate analyses. PCA generated new variables that summarize the variance in the original 18 independent morphometric variables described in Table 2. The HCA was used to summarize the position of the accessions relative to one another by sorting them into distinct groups. The unweighted pair group method using arithmetic average (UPGMA) was used for clustering the coffee genotypes.

Results

Morphological and agronomic diversity of local coffee populations

The current study provides a comprehensive characterization of 61 accessions of Arabica coffee from the main producing districts in Saudi Arabia. There were significant differences among accessions for all morphological attributes measured except for leaf width (Figures 2-5; Tables 3-4; Tables S1-S15). The variability was high for FL, FW, FT, FL/FW, BL, BW, BT, FFW, BDW, Flsh and Angl. It was moderate for BL/BW and LL/LW and low for FSph, BSph, LL and Inod.

Accessions KSA-3R, KSA-4R, KSA-12R, KSA-17R, KSA-20, KSA-35 and KSA-37 had compact canopies with a columnar shape (Figure 2). Most other accessions had open conical canopies. Most trees were 2.5 to 3 m tall except for accession KSA-9R that had a dwarf stature with short laterals and internodes and a very compact pyramidal canopy. Its trees had a height of about 1.5 m when they were 5 years old. Trees of accessions KSA-46 and KSA-47 were more than 4 m tall.



Figure 2. Illustration of variability in tree size and growth habit among coffee accessions collected from various sites in southwestern Saudi Arabia. KSA-3R (left) had a large columnar canopy, KSA-61 (center) had an open conical canopy while KSA-9R (right) had a pyramidal, dwarf and compact stature

The crotch angles of the 9-11th primaries with the main stem varied between 45 and 66° (Table S1). Accessions KSA-3R, KSA-7, KSA-11R, KSA-4R, KSA-8R, KSA15, KSA1R, KSA-45R, KSA-16R, KSA-30, KSA-69 and KSA-19R had relatively narrow crotch angles at the top of the canopy averaging 44.7°-46.7°. The crotch angles of accessions KSA-21, KSA-74, KSA-20, KSA-26, KSA-18R, KSA-9R, and KSA-12R varied from 61.7° to 66.0° indicating the tendency of their primaries to become plagiotropic at an early age.

The average length of the 2-6th internodes on four main plagiotropic primaries from the lower tiers of the tree was between 4.9 and 7.6 cm (Table S2). Accessions KSA-9R, KSA-6R, KSA-20R, KSA-10R, KSA-10, KSA-61, KSA-61, KSA-12, KSA-22, and KSA-30 had the shortest internodes that varied from 2.0 to 5.3 cm in length indicating the compactness of their canopies. The internodes of the dwarf KSA-9R measured 2.0±0.1

cm. The length of the internodes of accessions KSA-68, KSA-69, KSA-74, KSA-25, KSA-49, KSA-32, KSA-11, KSA-20 and KSA 67 varied from 6.9 to 7.6 cm due to their mostly open canopies.

The leaves of most accessions were elliptic with apiculate apices and undulated edges (Figure 4). However, the accessions varied slightly in leaf length and length to width ratio (LL/LW) but not in LW (Table S3-S5). Leaf length varied from 9.0 to 15.4 cm with accessions KSA-5R, KSA-10R, KSA-4R, KSA-42, KSA-9R, and KSA-7R having the shortest leaves while accessions KSA-29, KSA-31, KSA-32, KSA-35 and KSA-26 having the longest leaves. Finally, accessions KSA-45R, KSA-6R, KSA-15R, KSA-64, KSA-9R and KSA-4R had large LL/LW ratios ranging from 2.5 to 2.7 while KSA26, KSA-38, KSA-20, KSA-33, KSA-34 and KSA-32 had low ratios. The leaves of KSA-9R were mostly flat and less pointed (acute apex) compared to the other accessions (Figure 3).



Figure 3. Visual of the super compact and dwarf genotype KSA-9R. The new leaves were light bronze (left) and the cherries were medium to large (right). The tree produces year-around

The new leaves (growth flushes) of accessions KSA-7, KSA-42, KSA-37, KSA-31, KSA-18R, KSA-20, KSA-22, KSA-16R, KSA-12R, KSA-11R, KSA-44, KSA-43, KSA-6R, KSA-66, KSA-23, KSA-3R, KSA-35 and KSA-30 were green (Figure 4; Table S6); this group included genotypes that growers identified as either Kholani or Balady. Other accessions such as KSA-10, KSA-21, KSA-29, KSA-41, KSA-52 and KSA-61 had bronze young leaves, whereas, accessions KSA-8, KSA-19R, KSA-60 and KSA-69 had dark bronze flushes.

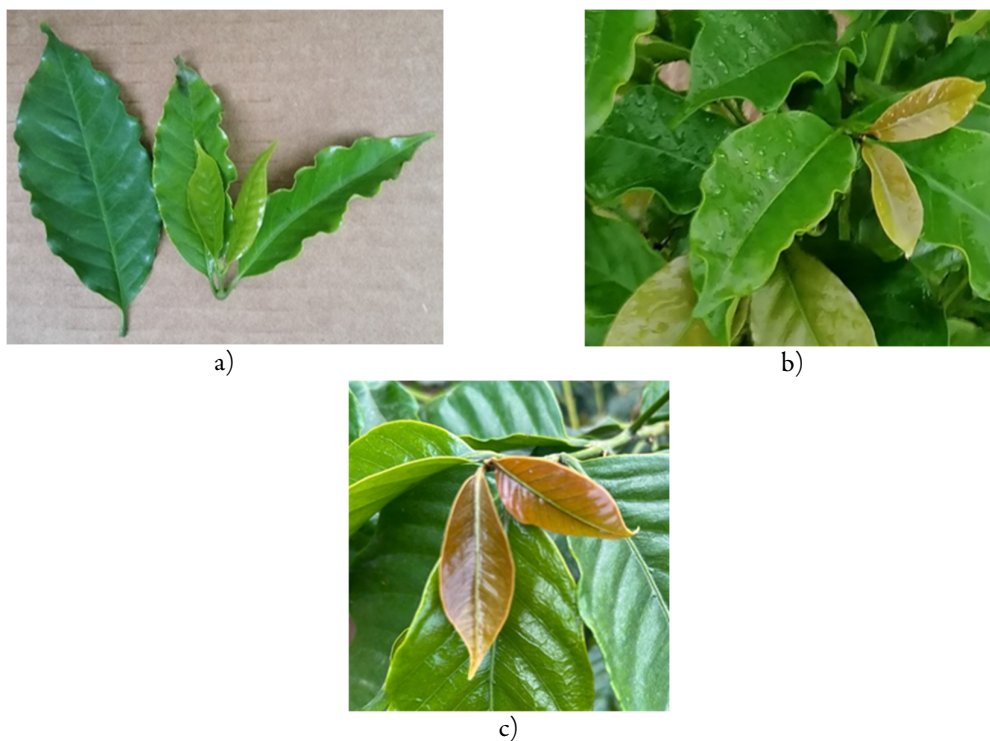


Figure 4. Examples of variability in the color of new growth flushes and mature leaves: new leaves of accessions KSA-20, KSA-30 and KSA43 were green (a), those of accessions KSA-10, KSA-29 and KSA-61 were bronze (b), whereas new leaves of accessions KSA-8, KSA-60 and KSA-69 were dark bronze (c)

Similarly, the cherries varied in size and shape among the coffee accessions (Figure 5; Table 5; Table S7-S11). The 100-cherry fresh mass varied from 76.3 to 234.8 g. Accessions KSA-7, KSA-8R, KSA-74 and KSA-28 had the lightest cherries whereas, KSA-52, KSA-29, KSA-25, KSA-38 and KSA-69 had the heaviest cherries. Cherry length varied from 13.5 to 18.0 mm (Table S7). Accessions KSA-64, KSA-9R, KSA-25 and KSA-12R had the shortest cherries whereas, KSA-69, KSA-52, KSA-33, KSA-62, KSA-29 and KSA-49 had the longest cherries. Accessions KSA-21, KSA-34, KSA-63 and KSA-74 had elliptic, sometimes, oblong cherries with pointed ends; the color of their cherries went from green to yellow then orange red as they matured. Their sphericity coefficient ranged from 0.81 to 0.83 and their length to width ratio ranged from 1.23 to 1.29 (Table S11). On the other hand, other genotypes such as KSA-20, KSA-29, KSA-31, KSA-38, KSA-52 and KSA-69 had oval bold-ended cherries with dull dark red color on a green background. Cherries of accessions KSA-44, KSA-4R, KSA 24, KSA-11R, KSA-10R, KSA-2R, KSA-20R, KSA-8R, KSA-37, KSA-3R, and KSA-1R were the most spherical (roundish); their sphericity coefficient ranged from 0.92 to 0.95 and their cherry length to width ratio ranged from 1.02 to 1.08.

Bean dry mass varied from 9.3 to 22.5 g in 100 beans (Table 4). Accessions KSA-66, KSA-49, KSA-10, KSA-21, KSA-62, KSA-7, KSA-31, KSA-26, KSA-38, KSA-8, KSA-69 and KSA-52 had the heaviest beans; their 100-bean dry mass varied from 18 to 22.5 g.



Figure 5. Illustration of variability in cherry and bean shape and size among coffee accessions collected from various sites in southwestern Saudi Arabia. Cherries and beans of KSA-21 (left) were elongated and pointed, those of KSA-52 (center) were oval and bold, while those of KSA-1R (right) were small and round (circular)

Table 3. Ranking of 61 local *C. arabica* accessions collected from southwestern Saudi Arabia according to cherry fresh mass (g/100 cherries)

Accession	Cherry fresh mass (g/100 cherries)
KSA-52	214.4 a
KSA-29	214.2 a
KSA-25	214.0 a
KSA-38	208.9 a
KSA-69	207.7 a
KSA-49	202.0 a
KSA-1R	200.0 a
KSA-26	197.4 a
KSA-4R	195.6 a
KSA-24	195.5 a
KSA-27	193.4 a
KSA-62	189.2 a
KSA-5R	186.4 b
KSA-33	186.2 b
KSA-10	185.6 b
KSA-20R	185.4 b
KSA-60	183.4 b
KSA-17R	183.0 b
KSA-20	182.0 b
KSA-61	179.4 b
KSA-59	178.7 b
KSA-63	176.1 b
KSA-16R	176.0 b

KSA-32	174.3 b
KSA-31	174.2 b
KSA-10R	173.4 b
KSA-66	173.4 b
KSA-15R	173.0 b
KSA-45R	172.7 b
KSA-30	172.2 b
KSA-19R	171.9 b
KSA-9R	168.0 b
KSA-3R	167.0 b
KSA-2R	165.0 b
KSA-11R	162.4 b
KSA-8R	159.0 b
KSA-21	157.9 b
KSA-67	156.0 c
KSA-68	154.3 c
KSA-39	153.9 c
KSA-37	152.1 c
KSA-34	146.6 c
KSA-12R	145.1 c
KSA-6R	144.6 c
KSA-41	143.7 c
KSA-65	141.2 c
KSA-44	141.1 c
KSA-70	141.0 c
KSA-47	137.0 c
KSA-22	133.3 c
KSA-18R	132.7 c
KSA-42	130.8 c
KSA-23	128.0 c
KSA-35	126.2 c
KSA-46	125.0 c
KSA-43	118.3 c
KSA-64	117.6 c
KSA-28	96.5 d
KSA-74	96.4 d
KSA-8	87.5 d
KSA-7	76.3 d

Mean separation by the Scott-Knott Effect Size Difference test at $P < 0.05$. Means that do not share the same letter are different

Table 4. Ranking of 61 local *C. arabica* accessions collected from southwestern Saudi Arabia according to bean dry mass (g/100 beans)

Accession	Bean dry mass (g/100 beans).
KSA-69	22.00 a
KSA-8	21.72 a
KSA-26	20.46 a
KSA-52	20.39 a
KSA-49	19.60 a
KSA-38	19.45 a
KSA-7	19.08 a
KSA-31	18.73 a
KSA-21	18.42 b
KSA-10	18.30 b
KSA-30	18.17 b
KSA-62	18.03 b
KSA-16R	17.90 b
KSA-9R	17.79 b
KSA-6R	17.60 b
KSA-25	17.46 b
KSA-27	17.40 b
KSA-60	17.37 b
KSA-61	17.34 b
KSA-28	16.70 b
KSA-32	16.70 b
KSA-59	16.64 b
KSA-66	16.59 b
KSA-24	16.50 b
KSA-4R	16.40 b
KSA-47	16.19 b
KSA-10R	16.00 b
KSA-33	15.96 b
KSA-15R	15.86 b
KSA-29	15.80 b
KSA-3R	15.78 b
KSA-20R	15.56 c
KSA-74	15.43 c
KSA-68	15.40 c
KSA-5R	15.10 c
KSA-39	15.07 c
KSA-34	14.99 c
KSA-46	14.77 c
KSA-45R	14.71 c
KSA-63	14.54 c
KSA-2R	14.40 c
KSA-65	14.28 c
KSA-8R	14.20 c

KSA-20	13.95 c
KSA-12R	13.80 c
KSA-17R	13.68 c
KSA-11R	13.50 c
KSA-44	13.29 d
KSA-35	13.26 d
KSA-67	13.17 d
KSA-37	13.09 d
KSA-23	12.80 d
KSA-19R	12.80 d
KSA-70	12.70 d
KSA-43	12.18 d
KSA-42	11.97 d
KSA-41	11.73 d
KSA-1R	11.00 d
KSA-64	10.95 d
KSA-18R	10.80 d
KSA-22	10.68 d

Mean separation by the Scott-Knott Effect Size Difference test at $P < 0.05$. Means that do not share the same letter are different

There were significant differences among the genotypes in bean length, width and length (Figure 5; Table S12-S14). Bean length varied from 7.9 mm to 11.2 mm, BW varied from 5.9 to 8.2 mm and BT varied from 3.1 mm to 4.5 mm. Accessions KSA-11R, KSA-12R, KSA-1R, KSA-67, KSA-6R, KSA-18R and KSA-64 had the shortest beans, while accessions KSA-69, KSA-52, KSA-38, KSA-37 and KSA-29 had the longest beans. Accessions KSA-12R, KSA-41, and KSA-6R had the narrowest beans while KSA-8, KSA-37, KSA-27, KSA-7R, KSA-38 and KSA-69 had the widest and thickest beans. Differences in bean shape were less obvious (Table S15, S16). In fact, most accessions had oval beans with length to width ratios between 1.21 and 1.52 and sphericity values between 0.62 and 0.72. However, accessions KSA-67, KSA-8, KSA-7, KSA-1R, KSA-49, KSA-11R, KSA-66, KSA-22 and KSA-68 had round (circular) beans; their length to width ratio ranged from 1.21 to 1.31 and their sphericity coefficient varied from 0.69 to 0.72. The exception was the beans of accessions KSA-21, KSA-34, KSA-61, and KSA-74 that were easily distinguishable by their elongated form; their length to width ratio was 1.46 ± 0.04 and their sphericity was 0.64 ± 0.01 .

Principal component analysis (PCA)

Eighteen morphometric and agronomic quantitative descriptors were used to characterize the diversity among 61 coffee accessions from southwestern Saudi Arabia using PCA and HCA. Generally, there were positive and significant correlations between cherry and bean geometric dimensions and mass (Table 5). The correlation of cherry length with width, thickness and fresh mass was +0.72, +0.70 and +0.56, respectively. The correlation of bean length with width, thickness and fresh mass was +0.74, +0.73 and +0.62, respectively. The correlation between cherry fresh mass and bean dry mass was only +0.42 indicating major differences in out-turn of green coffee among the accessions. Out-turn refers here to the percentage of dry mass of clean (green) beans to fresh mass of cherries; it varied from 11.0% to 34.6% with an overall average of 17.7%.

Table 5. Pearson’s correlation matrix for coffee tree quantitative morphometric and agronomic features considered in the study

	FL	FW	FT	FL/FW	FSph	BL	BW	BT	BL/BW	BSph	LL	LW	LL/LW	FFW	BDW	Flsh	Angl	Inod
FL	1.00																	
FW	0.72	1.00																
FT	0.70	0.88	1.00															
FL/FW	0.34	-0.40	0.26	1.00														
FSph	-0.35	0.27	0.37	-0.84	1.00													
BL	0.47	0.28	0.30	0.22	-0.20	1.00												
BW	0.23	0.20	0.26	0.02	0.01	0.74	1.00											
BT	0.33	0.29	0.34	0.03	0.02	0.73	0.85	1.00										
BL/BW	0.39	0.14	0.10	0.31	-0.31	0.54	-0.16	0.00	1.00									
BSph	-0.27	-0.04	0.00	-0.29	0.32	-0.45	0.19	0.23	-0.92	1.00								
LL	0.24	0.18	0.20	0.07	-0.07	0.20	0.01	0.10	0.28	-0.19	1.00							
LW	0.16	0.13	0.16	0.03	0.00	0.03	-0.06	0.04	0.10	-0.03	0.60	1.00						
LL/LW	-0.27	-0.10	0.19	-0.20	0.10	-0.15	-0.18	0.23	0.01	-0.08	-0.12	-0.51	1.00					
FFW	0.56	0.65	0.59	-0.14	0.06	0.34	0.20	0.33	0.22	-0.10	0.12	0.07	-0.03	1.00				
BDW	0.47	0.21	0.26	0.34	-0.30	0.62	0.57	0.59	0.20	-0.11	0.10	0.05	-0.16	0.42	1.00			
Flsh	0.31	0.24	0.25	0.08	-0.09	0.28	0.26	0.27	0.09	-0.04	0.07	-0.04	0.04	0.21	0.26	1.00		
Angl	-0.03	-0.21	0.18	0.24	-0.18	-0.05	-0.16	-0.15	-0.13	-0.14	0.13	0.04	-0.06	-0.10	-0.07	-0.07	1.00	
Inod	0.23	0.24	0.25	-0.05	0.03	0.06	-0.01	0.04	0.10	-0.05	0.29	0.26	-0.22	0.01	-0.01	0.14	0.02	1.00

The description of the labels and the unit of each trait are given in Table 2. In bold type are the coefficients that were statistically significant at $P < 0.05$

The first four principal components accounted for 71.9% of the variance (Table 6). These components explained 30.1%, 18.3%, 13.7% and 9.9% of the variance, respectively. We defined eigenvalues equal to or above 0.30 as significant and their associated parameters define the axis (Table 7). The relative discriminating power of the PCs as reflected by the eigenvalues, was highest in PC1. This axis represented mainly the geometric dimensions and the mass of the berries and beans (Table 7, Figure 6). PC2 reflected the shape of the berries and beans, measured by the ratio of the longitudinal diameter to the equatorial diameter, and the sphericity. The longitudinal diameter was positively correlated with the ratio of the longitudinal diameter to the equatorial diameter and negatively correlated with the sphericity of the berries and beans. The variables representing the leaf traits had little influence on PC1 and PC2 as they appear close to the center of the PCA score plot. PC3 reflected the dimensions of the leaves and the internodes that were segregated together on the loading plot and made a strong contribution to this component. As expected, leaf length-to-width ratio was negatively correlated with leaf width. PC4 represented leaf length-to-width ratio. New flush coloration did not have any significant contribution to any of the four axes. The highest contribution to the over-all variation among the accessions was that of fruit sphericity (FSph, eigenvalue = 0.47), leaf length-to-width ratio (-0.469), fruit length-to-width ratio (0.461), leaf width (0.42) followed by bean sphericity (0.384) (Table 7).

Table 6. Eigenvalues of the correlation matrix and the contribution of the first six variables to the variance

Variable	Eigenvalue	Percentage of variance (%)	Cumulative variance (%)
1	5.413	30.1	30.1
2	3.284	18.3	48.3
3	2.471	13.7	62.1
4	1.778	9.9	71.9
5	1.104	6.1	78.1
6	0.935	5.2	83.3

Table 7. Extracted eigenvectors of the first four principal components of eighteen morphological characters. In bold type are the large contributions of individual variables to each axis

Variable	Coefficients of PC1	Coefficients of PC2	Coefficients of PC3	Coefficients of PC4
FL	0.365	-0.066	0.134	-0.110
FW	0.292	0.270	0.218	-0.252
FT	0.309	0.260	0.205	-0.142
FL/FW	0.086	-0.461	-0.122	0.195
FSph	-0.090	0.470	0.119	-0.079
BL	0.350	-0.098	-0.228	-0.015
BW	0.269	0.132	-0.376	0.214
BT	0.308	0.147	-0.304	0.213
BL/BW	0.192	-0.358	0.150	-0.326
BSph	-0.114	0.384	-0.139	0.370
LL	0.163	-0.092	0.376	0.239
LW	0.136	-0.014	0.420	0.391
LL/LW	-0.170	0.013	-0.136	-0.469
FW _t	0.331	0.107	-0.032	-0.197
bDW	0.309	-0.080	-0.262	0.073
Flush	0.177	0.021	-0.092	-0.113
crotch	-0.053	-0.265	0.074	0.135
ind	0.133	0.034	0.348	0.123

The biplot of PC1 and PC2 (Figure 6) revealed that KSA-29, KSA-33, KSA-38, KSA-52, KSA-60 and KSA-69 were distinguished by the geometric dimensions of their berries and beans whereas, KSA-21, KSA-34, KSA-63, KSA-74 by cherry and bean length-to-width ratios. The first group had large and heavy cherries and beans while the second group had elliptic pointed cherries and beans. Accessions KSA-21, KSA-34, KSA-63 and KSA-74 clustered along PC2 indicating how strongly they were marked by the elongated shape of their fruit and beans. Overall, accessions KSA-4R, KSA-5R, KSA-8R, KSA-15R, KSA-24, KSA-27, KSA-38, KSA-33, KSA-45R, KSA-49, KSA-61, and KSA-62 were the most divergent in the group as they were associated with the largest eigenvalues in PC1. Accessions KSA-1R, KSA-2R, KSA-3R, KSA-4R, KSA-5R, KSA-8R, KSA-10R, KSA-11R, KSA-19R, KSA-61 and KSA-67 were spread along the PC2 axis indicating that they were distinguished by the sphericity of their berries and beans that were round. Accessions KSA-11R, KSA-18R, KSA-20 and KSA-24 were spread along PC3 axis indicative of their relatively long and wide leaves; KSA-9R was on the opposite side because of its narrower leaves. The variables are described in Table 2.

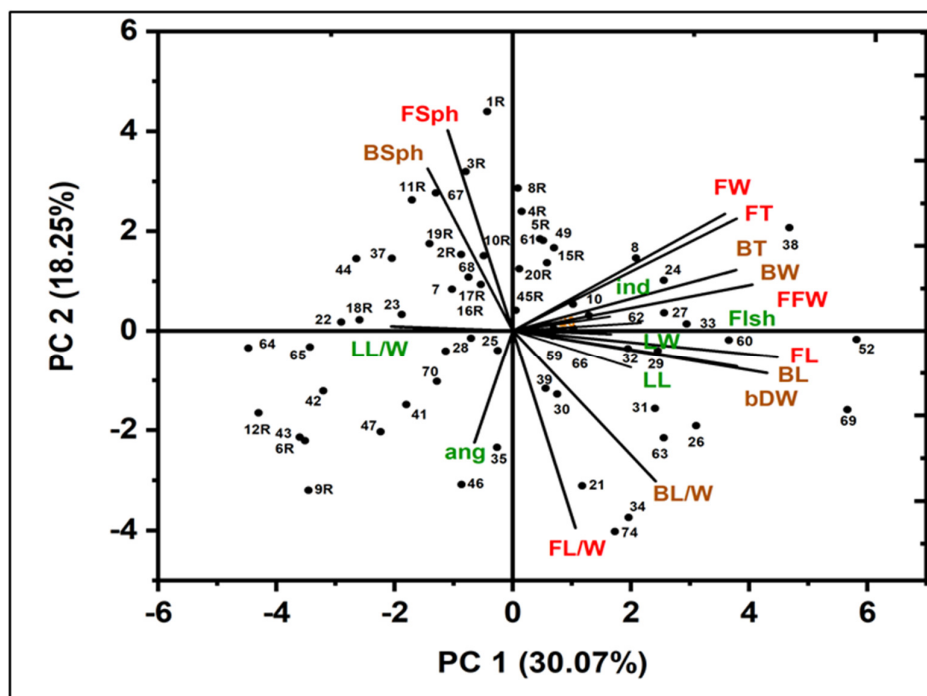


Figure 6. The biplot of the first two components resulting from a PCA of the variability in morphometric and agronomic characteristics of 61 coffee accessions from southwestern Saudi Arabia using 18 quantitative traits

The HCA clustered the accessions into three major groups (Groups I, II and III) in addition to accessions KSA-8, KSA-9R, KSA-11R, KSA-28 and KSA-37 which did not belong to any of the groups (Figure 7, Table 8). Group I contained two subgroups (subgroups 1 and 2) in addition to accession KSA1R. Subgroup 1 was heterogeneous and contained accessions from the Jazan region, Mahayel Assir and Jebel Shada Al-Aala; These accessions were KSA-2R, KSA-17R, KSA-20R, KSA-10R, KSA-5R, KSA-15R, KSA-8R, KSA-10, KSA-61, KSA-62, KSA-3R, KSA-4R, KSA-67 and KSA-45R. The trees of this subgroup were productive, compact and conical in shape. Some local growers refer to some members of this group as “Touffahi” for their large, roundish cherries. This group included the Shadawi cultivar (KSA-61) from Shada Al-Aala and the low caffeine KSA-10 from Mahayel Assir. They had a low percentage of elephant beans and peaberries (data not shown). Subgroup 2 consisted of four accessions from the Jazan region namely, KSA-16R, KSA-25, KSA-39, KSA-7R. They had medium sized cherries and large beans. The new leaves were green or light bronze. Group II contained Subgroup 3 which consisted mainly of old trees that growers refer to as “Balady” to mean old local genotypes; they included accessions KSA-22, KSA-44 and KSA-70 from Jebel Fayfa, KSA-46 and KSA-47 from Al-Gahr and KSA-64 and KSA-65 from Shada Al-Asfel. This subgroup is also heterogeneous, but most genotypes generally had vigorous and productive tall trees. Their new leaves were green or brown green. Their cherries and beans were small and had an intermediate shape. Group III included accessions mostly from the Jazan region; it can be subdivided into three subgroups (4, 5 and 6). Subgroup 4 included accessions KSA-20, KSA-24, KSA-26, KSA-27, KSA-30, KSA-31, KSA-32, KSA-35, KSA-59 and KSA-66. These genotypes represent the productive local varietal locally referred to as Kholani which is common in Jazan especially, in Jebel Fayfa and the villages of Al-Zoghli and Al-Gatil. The trees of these accessions had columnar canopies with bushy flat tops. The cherries were of medium size, oval in shape and had bold ends. The beans were elliptic and medium in size. They had a high percentage of peaberries (data not shown). Members of Subgroup 5, which included genotypes KSA-29, KSA-60, KSA-52, KSA-69, KSA-33 and KSA-38, had conical trees which bear large cherries and beans and had bronze young leaves. They had a low percentage of elephant beans and

peaberries. They were all from the Jazan region except for KSA-60 which was from Al-Majarda district of the Assir region. Subgroup 6 included accessions KSA-21, KSA-34, KSA-63 and KSA-74; they had conical trees with open canopies and long internodes. Their new leaves were bronze. The cherries were elliptic with apiculate apices and were medium to large. At maturity, they turned yellow then took a vivid orange red color. The beans were oval of medium size and had more than 10% of peaberries. There was only about two dozen trees of this type in Jebel Fayfa and Jebel Shada. The range of the eighteen measured parameters for each subgroup are given Table S17.

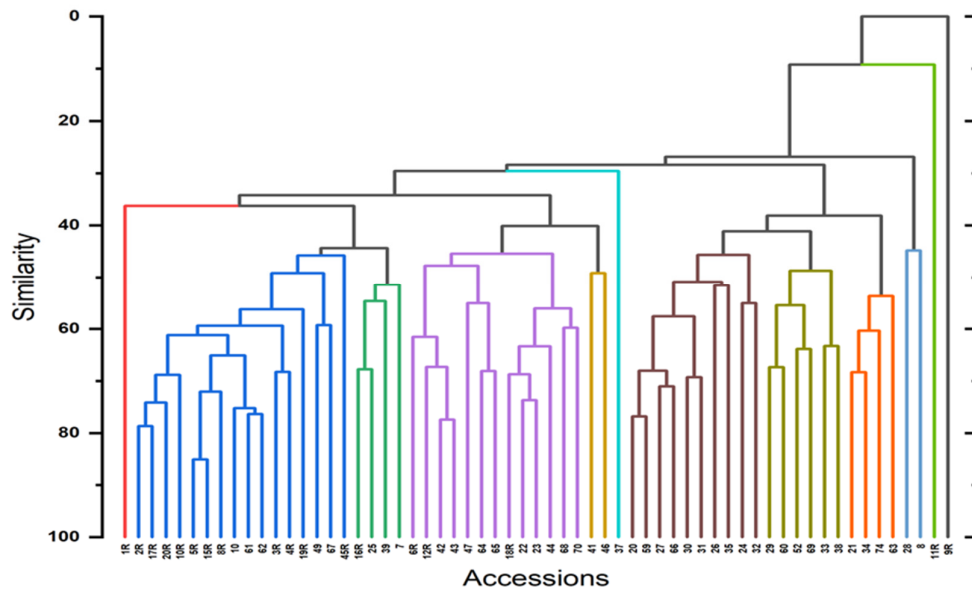


Figure 7. A cluster dendrogram illustrating the diversity among 61 Arabica coffee accessions from southwestern Saudi Arabia based on their morphometric features. The group average method was used for clustering using Euclidean distances. Accessions with 40% similarity or more were considered to belong to the same group

Table 8. Grouping of the 61 coffee accessions based on morphometric and agronomic features analyzed by PCA and HCA and confirmed by field observations

Group	Subgroup	Accessions in the subgroup	Notes
I	1 Shadawi and Touffahi type subgroup	KSA-2R, KSA-17R, KSA-20R, KSA-10R, KSA-5R, KSA-15R, KSA-8R, KSA-10, KSA-61, KSA-62, KSA-3R, KSA-4R, KSA-67, KSA-45R	Heterogeneous group from the Jazan, Assir and Al-Baha regions. The trees were productive, compact and pyramidal in shape. Some local growers refer to some members of this group as “Touffahi” for their large, round berries. This group includes the Shadawi cultivar (KSA-61) from Shada Al-Aala and the low caffeine KSA-10 from Mahayel Assir.
	2	KSA-16R, KSA-25, KSA-39, KSA-7	Accessions from the Jazan region. They had medium size cherries and large beans. The new leaves were green or light bronze.
II	3 Balady subgroup	KSA-6R, KSA-42, KSA-43, KSA-47, KSA-64, KSA-65, KSA-18R, KSA-22, KSA-23, KSA-44, KSA-68, KSA-70, KSA-41, KSA-46	Productive large trees with green or light bronze leaves and small beans. The cherries and beans had an intermediate shape. Growers referred to these old genotypes as “Balady”. They were found in all three regions.

III	4 Kholani-type subgroup	KSA-20, KSA-59, KSA-27, KSA-66, KSA-30, KSA-31, KSA-26, KSA-35, KSA-24, KSA-32	Kholani types were found in Jebel Fayfa, Al-Zoghli and Al-Gatil. The trees had columnar canopies with bushy flat tops. The cherries and beans were medium in size. The beans were oval.
	5 "Jumbo"-cherry subgroup	KSA-29, KSA-60, KSA-52, KSA-69, KSA-33, KSA-38	They had large bold cherries. The beans were oval and large. The new leaves were bronze. The trees were productive.
	6 Elongated-cherry subgroup	KSA-21, KSA-34, KSA-74, KSA-63	They had long, pointed, medium-size berries and beans. The new leaves were bronze. The trees had open canopies with long internodes. There were only a limited number of trees of this type in Jebel Fayfa and Jebel Shada.
Accessions not belonging to any group		KSA-1R, KSA-8, KSA-9R, KSA-11R, KSA-28, KSA-37	KSA-9R was a compact dwarf that produced year around.

Discussion

The analysis of variance had revealed considerable variability among the Arabica populations studies for most of the morphology traits considered. Part of the variability was likely due to the influence of environmental factors such as rainfall and soil fertility and differences in cultural practices. Nevertheless, most of the variance appears to be due to genetic differences since there were measurable differences even between genotypes collected from the same garden. For instance, the eighteen accessions from the small germplasm collection in Jebel Fayfa (JCGR) (KSA-1R-20R and KSA-45R) were planted in a small plot with a uniform soil and received the same care; still, they displayed considerable differences in tree size, growth habit and cherry and bean size and shape. Similarly, if we consider genotypes KSA-20 and KSA-21, KSA-34 and KSA-35, KSA-41 and KSA-42, KSA-59 and KSA-60, and KSA-61 and KSA-63, each two accessions came from the same small garden, yet they differed clearly in most morphometric traits (Figures 6-7, Figures S1-S16). Tounekti *et al.* (2017) carried out a similar study on 19 accessions from the same area and reported large differences in most morphometric traits they measured. Such variability in several interesting traits especially those contributing to tree yield and bean quality is important and can be the basis of breeding programs to develop new improved varieties through crossing and selection.

The existence of considerable genetic diversity among local Arabica coffee populations was also confirmed by previous genetic studies on Saudi (Al-Ghamedi *et al.*, 2023) and Yemeni (Montagnon *et al.*, 2021; Montagnon *et al.*, 2022b) cultivars. Montagnon *et al.* (2021) and Eskes and Mukred (1990) reported that most Yemeni coffee cultivars arised from ancient "heirloom" populations of *C. arabica* first naturalized hundreds of years ago. The results of the present study and several others that delt with genetic variability in Arabica coffee populations in the KSA and Yemen (Eskes and Mukred, 1990; Tounekti *et al.*, 2017; Montagnon *et al.*, 2021) support the hypothesis that the Arabian Peninsula represents the most important center of coffee diversity outside the species' center of origin in Ethiopia and South Sudan.

The variability among Saudi coffee populations may be due to either evolutionary trends, natural mutations occurring within these populations (Anthony *et al.*, 2001; Lachermes *et al.*, 2009) or original diversity due to multiple introductions of genetic material from various locations in eastern Africa over an extended period of time (Montagnon *et al.*, 2022a; Al-Ghamedi *et al.*, 2023). The lack of consistency in the magnitude of certain traits such as growth flush color, cherry shape, internode length, crotch angle and intensity of ramification of the primaries among members of the same subgroup as identified by multivariate analysis of morphometric data and SRAP data (Al-Ghamedi *et al.*, 2023) suggests that either these populations are still

segregating or there was a strong influence of environmental factors such as rainfall, soil, and elevation on the phenotypes (Ghafoor *et al.*, 2003; Cheng *et al.*, 2016; Velásquez and Banchón, 2022).

The characteristics of 'Typica' variety seemed to prevail in the districts of Almajarda, Mahayel Assir and Shadha Al-Asfel while those of 'Bourbon' were more evident in Jebel Fayfa, Eddayer, Tallan and Al-Gahr. Accessions KSA-45R, KSA-6R, KSA-65, KSA-60 seemed to belong to the first group while accessions KSA-32, KSA-38, KSA-33, KSA-20, KSA-7 and KSA-21 seemed to belong to the second. Similar results were reported by Tounekti *et al.* (2017).

Based on our observations and information shared by elder growers, Kholani-type populations were the most widely planted in the Jazan region while Shadawi was the main cultivar in the Al-Baha region. Representative accessions of the Kholani-type are KSA-20, KSA-26, KSA-30 and KSA-35. Shadawi was represented by accessions KSA-61 and KSA-62. Both cultivars are productive and give a high-quality liquor and much appreciated by the local consumer (Tounekti *et al.*, 2017; Kathurima *et al.*, 2022).

Based on HCA grouping and field observations, KSA-2R, KSA17R and KSA-20R were remarkably similar and may be synonyms. The same thing can be said for 5R and 15R, KSA-61 and KSA-62, KSA-20 and KSA-59 and KSA-64 and KSA-65.

Previous studies using morphometric traits (Kebede and Bellachew, 2008; Kathurima *et al.*, 2022) show that it is often difficult to differentiate coffee genotypes based only on a limited number of traits. Therefore, we included 18 traits relative to tree canopy structure, fruit and seed morphology, mass and color, leaf properties and internode length to identify the genotypes. In fact, most accessions had bold elliptic cherries and oval beans with length to width ratios between 1.12 and 1.18 for cherries and 1.36 to 1.41 for beans except for members of Subgroup 1 that had roundish (circular) cherries and beans and members of Subgroup 6 that had elongated fruit and beans. Despite the statistical differences in cherry and bean geometric dimensions, it was still hard to tell the accessions apart based on these descriptors alone. Furthermore, it appears from the present study that not all traits are worth considering for the analysis. For instance, anthocyanin coloration of new growth flushes is an easy feature to observe but it too does not seem to be a reliable descriptor for a different reason. Indeed, we can find within the same subgroup genotypes with green new leaves and others with bronze leaves. For instance, KSA-3R of Subgroup 1 (the Touffahi subgroup) had green flushes whereas KSA-10 and KSA-61 of the same subgroup had bronze flushes. Similarly, KSA-12R of Subgroup 5 (the Jumbo-cherry subgroup) had green flushes whereas KSA-60 of the same subgroup had dark bronze flushes. Besides, the contribution of this variable was weak in all first four principal components of the PCA; the coefficients of the variable 'flush color' in the extracted eigenvectors of the four principal components were only 0.18, 0.02, -0.09 and -0.11 for PC1, PC2, PC3 and PC4, respectively. Part of the reason could be that many of these populations are still segregating. Al-Ghamedi *et al.* (2023) studied this same population using SRAP markers and found that 25% of the genotypes have admixture ancestry. They also found that members of the same genetic group have growth flushes of different colors. For instance, they found that accessions KSA-3R, KSA-6R and KSA-44 which have green new leaves belonged to the same group (Subpopulation V in their study) as KSA-1R and KSA-21 which have bronze new leaves. Therefore, this descriptor is of low value in explaining the diversity of coffee genotypes and differentiating them.

Earlier studies suggest a strong influence of geographic origin on diversity among coffee genotypes (Ghafoor *et al.*, 2003; Wintgens, 2012; Kathurima *et al.*, 2022). However, the results of our study show that accessions of coffee harvested from the same valley were grouped into different clusters, suggesting a substantial genetic diversity within the populations of each region. The Tallan valley provides a clear illustration of this; four accessions growing at the same location, yet they were grouped into four different clusters.

The PCA revealed that the first four axes explained 71.9% of the total variability among the genotypes. The traits with the largest eigenvalues have the highest power of discrimination among the genotypes (Falconer and Mackay, 1996). Fruit and bean geometric dimensions and fruit fresh weight and bean dry weight were the most represented by PC1. PC2 represented cherry and bean form, i.e., sphericity and length-to-width ratio while PC3 summarized leaf dimensions and internode length while PC4 represented leaf length-to-width ratio.

Of all the parameters analyzed, those related to fruit and bean size and shape and leaf width and leaf length-to-width ratio had the highest power of discrimination, and were, therefore, the most useful for the description of Arabia coffee germplasm. This finding is partly in agreement with Olika *et al.* (2011) who found that bean length and 100-bean dry mass contributed the most to the variation among Limmu coffee accessions. In a similar study on coffee populations from Saudi Arabia, Tounekti *et al.* (2017) found that cherry and bean geometric proprieties in addition to tree productivity are the most important descriptors for the coffee tree.

In the current study, the HCA results grouped the Arabica coffee accessions into six clusters (subgroups) while six accessions did not belong to any group and remained separated. This clustering of the genotypes is similar to what was reported by Tounekti *et al.* (2017) even though they only studied 19 accessions. Al-Ghamedi *et al.* (2023) studied the genetic diversity of 56 of the 61 accessions using SRAP markers. The result of the clustering was somewhat different in the two studies although the majority of the members of Subgroups 3 and 4 in the present study are found in Subpopulation 3 of their study. Similarly, most members of Subgroups 1 and 2 in the present study are found in Subpopulations 1 and 2 of their study. Part of the discrepancy could be due to phenotypic differences due to genotype x environment interactions; in fact, the correlation between location and grouping based on morphology was 0.39 ($P = 0.006$). Falconer and Mackay (1996) stated that genetic diversity can arise through diversity in origin (geographical separation), ancestral relationship, gene frequencies and morphology. They indicated that plants differing in either one or more of these factors would differ by a significant number of genes.

Most of the inter-cluster distances were large indicating the presence of considerable variability that can be exploited through selection and hybridization as characters with high variability are expected to provide a high level of gene transfer during breeding programs (Hedrick, 2000). It is generally accepted that there is a higher chance to obtain transgressive segregation and enhanced heterosis when we cross accessions from distant genetic groups due to unique allelic differences (Ghaderi *et al.*, 1984; Hedrick, 2000; Tounekti *et al.*, 2018). These findings are significant because the area where coffee is grown in southwestern Saudi Arabia is arid to semi-arid (De Pauw, 2002), therefore, the collected germplasm could contain interesting traits of abiotic stress tolerance which can provide the base material for developing drought and heat tolerant cultivars (Tounekti *et al.*, 2018).

According to our results, the maximum recombination and segregation of the progenies is expected from crosses involving parents selected from the divergent genotypes not belonging to any group followed by those selected from Subgroups 3 and 5. Still, the breeder needs to choose the parent according to desired characters he is aiming for.

Morphological and pomological traits, especially those included in PC1 and PC2, allowed the effective classification of the 61 coffee accessions into six different clusters (subgroups) and should be considered in selecting parents for future crossing programs. It is safe to assume that, depending on the goals of the breeding program, each cluster has a unique set of breeding values that breeders can use to enhance the genetic diversity and conserve local coffee resources for future breeding programs.

Conclusions

The study revealed considerable morphological variability among the Arabica coffee populations in southwestern Saudi Arabia. These results were based on an uncontrolled field survey that should be complemented with a more detailed experimental study once a germplasm collection of these genotypes is established. Of all the morphometric parameters analyzed, those related to fruit and bean mass, size and shape and crotch angle (Angl) had the highest power of discrimination, and were, therefore, the most useful for the description of coffee germplasm. Accessions KSA-8R, KSA-4R, KSA-61, KSA-5R, KSA-49, KSA-45R, KSA-15R, KSA-24, KSA-62, KSA-24, KSA-38, KSA-27, and KSA-33 were the most divergent genotypes. These

accessions contributed the most to the diversity of the collection and should be considered for future coffee breeding programs.

Authors' Contributions

Conceptualization: HK, MM and TT; Data curation: HK, MM, TT, MA, MS, WA and DS; Formal analysis: HK, MO, FA, MS and MA; Funding acquisition: HK, MO, MM, TT, ZA and WA; Investigation: HK, TT, MA and DF; Methodology: HK, MO, FA and MA; Project administration: HK, MM and TT; Resources: HK, MO and DS; Supervision: HK and MM; Writing - original draft: HK, MO, MA and TT; Writing - review and editing: HK, MM, TT, MO, ZA, WA, MA, MS and DS.

All authors read and approved the final manuscript.

Ethical approval

Not applicable.

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Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

References

- Ahmed S, Brinkley S, Smith E, Sela A, Theisen M, Thibodeau C, ... Cash SB (2021). Climate change and coffee quality: systematic review on the effects of environmental and management variation on secondary metabolites and sensory attributes of *Coffea arabica* and *Coffea canephora*. *Frontiers in Plant Science* 12:1-20. <https://doi.org/10.3389/fpls.2021.708013>
- Al-Ghamedi K, Alaraidh I, Afzal M, Mahdhi M, Al-Faifi Z, Oteef MD, ... Khemira H (2023). Assessment of genetic diversity of local coffee populations in southwestern Saudi Arabia using SRAP markers. *Agronomy* 13:302. <https://doi.org/10.3390/agronomy13020302>
- Anthony F, Bertrand B, Quiros O, Wilches A, Lashermes P, Berthaud J, Charrier A (2001). Genetic diversity of wild coffee (*Coffea arabica* L.) using molecular markers. *Euphytica* 118:53-65. <https://doi.org/10.1023/A:1004013815166>
- Cheng B, Furtado A, Smyth HE, Henry RJ (2016). Influence of genotype and environment on coffee quality. *Trends Food Science and Technology* 57:20-30. <https://doi.org/10.1016/j.tifs.2016.09.003>
- Davis AP, Chadburn H, Moat J, O'Sullivan R, Hargreaves S, Nic Lughadha E (2019). High extinction risk for wild coffee species and implications for coffee sector sustainability. *Science Advances* 5:eaav3473. <https://doi.org/10.1126/sciadv.aav3473>
- De Pauw E (2002). An agroecological exploration of the Arabian Peninsula. International Center for Agricultural Research in the Dry Areas (ICARDA), Beirut, Lebanon.

- Eskes A, Mukred A (1990). Coffee survey in PDR Yemen. In: Proceeding of the 13th International Scientific Colloquium on Coffee. Association Scientifique Internationale pour le Café pp 582-590.
- Eulefeld G (1979). The UNESCO-UNEP program in environmental education. European Journal of Science and Education 1:113-118.
- Falconer DS, Mackay TC (1996). Introduction to quantitative genetics. Pearson Education (4th ed), Harlow, UK.
- Ferreira T, Shuler J, Guimarães R, Farah A (2019). Introduction to coffee plant and genetics. In: Farah A (Ed). Coffee: Production, quality and chemistry. The Royal Society of Chemistry, London, UK pp 1-25.
- Gennari P, Heyman A, Kainu M (2015). FAO statistical pocketbook. World Food and agriculture. Food and Agriculture Organization of the United Nations, Rome, Italy.
- Ghaderi A, Adams MW, Nassib AM (1984). Relationship between genetic distance and heterosis for yield and morphological traits in dry edible bean and fava bean. Crop Science 24:37-42. <https://doi.org/10.2135/cropsci1984.0011183X002400010009x>
- Ghafoor A, Ahmad Z, Hashmi NI, Bashir M (2003). Genetic diversity based on agronomic traits and SDS-PAGE markers in relation to geographic pattern of blackgram (*Vigna mungo* (L.) Hepper). Journal of Genetics and Breeding 57:5-14.
- Hedrick PW (2000). Genetics of populations. Jones and Bartlett Publishers (2nd ed), Burlington, MA, United States.
- IPGRI (1996). Descriptors for coffee (*Coffea* spp. and *Psilanthus* spp.). Biodiversity International, Rome, Italy.
- Johnson RA, Wichern DW (2002). Applied multivariate statistical analysis. Prentice Hall (5th ed), Upper Saddle River, NJ, United States.
- Kathurima C, Ghosh K, Bandar A, Alwshigry A, Alojaimi A, Kimemia J (2022). Un-veiling physical and sensory quality of arabica coffee produced in the kingdom of Saudi Arabia. Food and Agriculture Organization of the United Nations, c/o Saudi Arabia's Ministry of Environment, Water and Agriculture: Riyadh, Saudi Arabia.
- Kebede M, Bellachew B (2008). Phenotypic diversity in the Hararge coffee (*Coffea arabica* L.) germplasm for quantitative traits. East African Journal of Science 2:13-18. <https://doi.org/10.4314/eajsci.v2i1.40358>
- Lachermes P, Bertrand B, Etienne H (2009). Breeding coffee (*Coffea arabica*) for sustainable production. In: Jain SM, Priyadarshan PM (Eds). Breeding plantation tree crops: Tropical species. Springer, New York, NY, United States pp 525-543.
- Montagnon C, Mahyoub A, Solano W, Sheibani F (2021). Unveiling a unique genetic diversity of cultivated *Coffea arabica* L. in its main domestication center: Yemen. Genetic Resources Crop Evolution 68: 2411-2422. <https://doi.org/10.1007/s10722-021-01139-y>
- Montagnon C, Rossi V, Guercio C, Sheibani F (2022b). Vernacular names and genetics of cultivated Coffee (*Coffea arabica*) in Yemen. Agronomy 12(8):1970. <https://doi.org/10.3390/agronomy12081970>
- Montagnon C, Sheibani F, Benti T, Daniel D, Bote AD (2022a). Deciphering early movements and domestication of *Coffea arabica* through a comprehensive genetic diversity study covering Ethiopia and Yemen. Agronomy 12:3203. <https://doi.org/10.3390/agronomy12123203>
- Olika K, Sentayehu A, Taye K, Weyessa G (2011). Variability of quantitative traits in limmu coffee (*Coffea arabica* L.) in Ethiopia. International Journal of Agricultural Research 6:482-493. <https://doi.org/10.3923/ijar.2011.482.493>
- Osorio N (2002). The global coffee crisis: a threat to sustainable development. ICO, London, UK.
- Sayed OH, Masrahi YS, Remesh M, Al-Ammari B (2019). Coffee production in southern Saudi Arabia highlands: current status and water conservation. Saudi Journal of Biological Science 26:1911-1914. <https://doi.org/10.1016/j.sjbs.2019.03.002>
- Stewart S, Ivy MA, Anslyn EV (2014). The use of principal component analysis and discriminant analysis in differential sensing routines. Chemical Society Reviews 43:70-84. <https://doi.org/10.1039/C3CS60183H>
- Tounekti T, Mahdhi M, Al-Turki T, Khemira H (2018). Water relations and photo-protection mechanisms during drought stress in four coffee (*Coffea arabica*) cultivars from southwestern Saudi Arabia. South African Journal of Botany 117:17-25. <https://doi.org/10.1016/j.sajb.2018.04.022>
- Tounekti T, Mahdhi M, Al-Turki TA, Khemira H (2017). Genetic diversity analysis of coffee (*Coffea arabica* L.) germplasm accessions growing in the southwestern Saudi Arabia using quantitative traits. Natural Resources 8:321-336. <https://doi.org/10.4236/nr.2017.85020>

Velásquez S, Banchón C (2022). Influence of pre-and post-harvest factors on the organoleptic and physicochemical quality of coffee: a short review. *Journal of Food Science and Technology* 60(10):1-13. <https://doi.org/10.1007/s13197-022-05569-z>

Wintgens JN (2012). Coffee bean quality assessment. In: Wintgens JN (Ed). *Coffee growing, processing, sustainable production: A guidebook for growers, processors, traders and researchers*. Wiley-VCH Verlag GmbH & Co. KGaA (2nd ed), Weinheim, Germany pp 818-828.



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