Moroccan Journal of Biology Number 13 (2016)

Contribution to the discussion about the national olive orchard heterogeneity through a morphological study of some olive trees (*Olea europaea* L.) cultivated in the Beni Tajjit site, SW of the Oriental Region of Morocco

E. Khlil, A. Mihammou, M. Abid, E. Tahri

Laboratoire de Biologie des Plantes et des Microorganismes, Département de Biologie, Faculté des Sciences, Université Mohamed Premier, Oujda, Morocco

Abstract

In this present study, we performed a morphological characterization of some olive trees cultivated under irrigated conditions in one geographically marginal site (Beni Tajjit) which is localized in the south-western part of the Moroccan Oriental Region. We used morphological traits to assess morphological diversity of these olive trees as well as to determine their phenotypic relationships with the main known olive Moroccan cultivars. A total of eighteen olive trees (noted from BT_1 to BT_{20}) selected for their apparent morphological differences were analysed by using 23 morphological characters (2 for leaves, 2 for inflorescences, 9 for fruits, and 10 for endocarps) belonging to those suggested by the International Olive Oil Council. Results revealed that differences between all studied olive trees were ranged from 1 (so be it 4.3%) as minimum to 9 (so be it 39.1%) as maximum characters. The dendrogram resulting from the UPGMA method based on morphological characters and using Squared Euclidean distance revealed five major groups of olive trees according to their aggregation distance values which were 1; 1.35; 1.7 and 5.9. Between these five groups as well inside of every group, differences interested both the number and the category of character traits. Otherwise, the 18 studied olive trees showed 2 to 16 morphological traits as difference with the most cultivated Moroccan olive varieties Picholine marocaine, Menara, Haouzia, Meslala, Dahbia, and Bouchouika. Results point clearly that the genetic diversity in Moroccan olive material could be much higher than what was assessed previously.

Keywords: Olea europaea L., Biodiversity, Olive varieties, Picholine marocaine, Morphological traits.

Abbreviations and acronyms: BT: Beni Tajjit site or olive tree from the Beni Tajjit site; IOOC: International Olive Oil Council;UPGMA: Unweighted pair-group method.

Introduction

Cultivated olive (Olea europaea L.) is known by its large adaptation to many varied environmental conditions and (Rugini et al., 2011). Mediterranean area represents not only 90% of the olive cultivated area worldwide but also 90% of the world olive production (Rugini et al., 2011). The primary producers of olive oil are Spain, Italy, Greece, Tunisia, Turkey, Syria, Morocco, and Portugal (FAOSTAT, 2006). According to Moroccan Ministry of agriculture data (Ministère de l'Agriculture et de la Pêche Maritime, 2013), the olive knows greater expansion sector a

considering as well as cultivated superficies than production. It has major socio-economical role. It contributes to decrease the flu of rural migration (Ouazzani et al., 1996). Its geographical distribution is also varied (FAO, 2009b): Mountainous area (20,000 ha representing 36% of the production); Rain fed area (100,000 ha representing 18% of the production); Irrigated area (220,000 ha representing 39% of production); and other marginal areas (40,000 ha representing 7% of the production).

At the world scale, olive includes a large number of varieties with significant phenotypic and genetic diverseness (Ziliotto et al., 2002; Idrissi & Ouazzani, 2004). More than 1200 cultivars were identified (FAO, 2009a). The classical approach in the identification of olive cultivars has been based on agronomical and/or morphological studies. It consists of measures such as production average, vigour, and precocity, etc. as well as the direct observation of several distinctive traits on leaves, inflorescences, fruits, endocarps, and branches (Barranco & Rallo, 1984; Barranco et al., 2000; Idrissi & Ouazzani, 2004; Poljuha et al., 2008). In this case, the International Olive Oil Council (IOOC) has considered a set of some morphological characters concerning notably tree, leaf, inflorescence, fruit and endocarp and then standardized morphological methods were usually applied for variety description and identification (COI. 1997). These descriptors named primary have been used for characterization and identification of a great number of olive cultivars in Spain (Barranco & Rallo, 1986; Tous & Romero, 1992), France (Ruby, 1916), and Tunisia (Msallem et al., 2000; Trigui & Msellem, 2002) for examples.

Discrimination of varieties based on morphology evaluation is limited by environmental conditions effect, the need for extensive observations of mature plants and requirement of well-trained staff (Belaj *et al.*, 2001). Occurrence of large number of varietal homonymy (varieties having the same name but that are genetically different) and synonymy (varieties having different names but that are genetically the same), and clonal selections have

Materials and methods Plant material and study site

This study was carried out during two growing seasons 2010-2011 and 2011-2012 and conducted on eighteen olive trees (noted from BT1 to BT20) among those which were cultivated in the Beni Tajjit complicated varietal identification and characterization in olive tree (Barranco et al., 2000; Bandelj et al., 2002; Therios, 2005). Therefore, more comprehensive studies using reliable markers were needed to have a better understanding of genetic diversity levels in olive cultivars, which be of use in the cultivars mav identification. Molecular techniques were precise also used to genetic their characterization owning to reliability reproducibility, and independence from environmental conditions (Poljuha 2008). et al., Molecular studies have started with the use of isoenzyme markers (Ouazzani et al., 1993; Trujillo et al., 1995) and later have been carried out using DNA markers as restriction fragment length polymorphisms (RFLP) (Besnard et al., 2001), random amplified polymorphic DNAs (RAPDs) (Gomes et al., 2008; Erfatpour et al., amplified fragment 2011). length polymorphisms (AFLPs) (Bandelj et al., 2004; Montemurro et al., 2005) and microsatellite markers (La Mantia et al., 2005; Baldoni et al., 2009; Shabanimofrad et al., 2011).

Our research tents to examine morphological characteristics of some olive trees planted in one sub-marginal site which is localized in the SW part of the Oriental Region of Morocco. This present work was conducted on a restricted number of olive trees that were chosen for their morphological differences observed previously in the field. Results would participate to obtain further information on the Moroccan olive orchards diversity, and later to identify the more adapted cultivars for different pedoclimatic conditions.

site (BT) that is localized in the south-west part of the oriental region of Morocco (Latitude: 32; Longitude: -3.4; Altitude: 1,100 meter of sea level). Olive trees present some visual differences in their morphological traits. They grew in the same pedoclimatic conditions under traditional agricultural practices with sufficient irrigation (gravitational system) during specially the non-rainfall periods. Local climate is arid with annual rainfall mean less than 200mm and with mild springs and hot and dry summers.

Morphological Characteristics

The olive descriptors used in this study were according to those of the IOOC standards (COI, 1997). Four organ types, leaves, inflorescences, fruits and endocarps were subjected to the observation. Twenty three morphological characters (2 of leaf, 2 of inflorescence, 9 of fruit and 10 of endocarp) that had a very high discriminating power for the identification of olive varieties were determined at less on 40 samples of each olives organ (See the list above, Table 1). The final category of each morphological character was then determined by considering the dominant categories only. For comparison between olives. quantitative variables corresponding to the character categories (numbers from 1 to 4, see Table 1) were the hierarchic classification used in (Unweighted pair-group method: UPGMA) analysis based on the squared Euclidean distance.

Table 1. List and abbreviation of morphological studied characters (Numbers in brackets correspond to the character categories).

Leaf parameters:	
Shape (LS): Elliptic (1), Elliptic-lanceolate (2), Lanceolate (3)	
Longitudinal curvature of the blade (LC): Epinastic (1), Flat (2), Hyponastic (3), Hélicodal (4)	
Inflorescence parameters:	
Length (IL): Short (1), Medium (2), Long (3)	
Number of flowers per inflorescence (IN): Scare (1), Medium (2), High (3)	
Fruit parameters:	
Shape (FS): Spherical (1), Oval (2), Elongated (3)	
Symmetry (FSy): Symmetrical (1), weakly asymmetrical (2), Asymmetrical (3)	
Position of maximum transverse diameter (FD): Towards base (1), Central (2), Towards apex (3)	
Apex (FA): Pointed (1), Rounded (2)	
Base (FB): Truncated (1), Rounded (2)	
Nipple (FN): Absent (1), weakly present (2), Evident (3)	
Presence of lenticels (FPL): Sparse (1), Numerous (2)	
Dimension of lenticels (FDL): Small (1), Large (2)	
Ripeness start (FR): From base (1), Uniform (2), From apex (3)	
Endocarp parameters:	
Shape (ES): Spherical (1), Oval (2), Elliptic (3), Elongated (4)	
Symmetry in position A (ESA): Symmetrical (1), weakly asymmetrical (2), Asymmetrical (3)	
Symmetry in position B (ESB): Symmetrical (1), weakly asymmetrical (2)	
Position of the maximum transverse diameter (ED): Towards base (1), Central (2), Towards apex (3)	
Apex (EA): Pointed (1), Rounded (2)	
Base (EB): Truncated (1), Pointed (2), Rounded (3)	
Surface (ESu): Smooth (1), Rugose (2), Scabrous (3)	
Number of fibrovascular grooves (ENG): Reduced (1), Medium (2), Elevated (3)	
Distribution of fibrovascular grooves (EDG): Uniform (1), Grouped around suture (2)	
Bill of apex (EM): Without mucro (1), with mucro (2)	

Results

Morphological characteristics of the olive trees from the Beni Tajjit site

The morphological characterization shows that the eighteen studied olive trees presented 11 same traits among the twenty three studied traits (Table 2). However, some character categories were observed in very restricted number of olive trees and sometimes in just one olive tree. This was the case of: (i) the leaf shape that is elongated in the BT14, BT15 and BT19 olive trees; (ii) the inflorescence with a medium length in the BT14 olive tree; (iii) fruit with a spherical shape in the BT13 and BT18 olive trees; (iv) endocarp with

elongated shape in the BT6 and BT9 olive trees, and (v) endocarp with smooth surface in the B16 olive tree.

Table 2. Quantitative variables corresponding to the morphological characteristic categories of the four olive organ types, leaves, inflorescences, fruits and endocarps that were subjected to the observation in the eighteen olive trees from the Beni Tajjit site.

	BT1	BT2	BT3	BT5	BT6	BT7	BT8	BT10	BT11	BT12	BT13	BT14	BT15	BT16	BT17	BT18	BT19	BT20
Leaves																		
LS	2	2	2	2	2	2	2	2	2	2	2	3	3	2	2	2	3	2
LC	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
Inflores	cence	s																
IL	1	1	1	1	1	1	1	1	1	1	1	2	1	1	1	1	1	1
IN	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
Fruits																		
FS	2	2	2	2	2	2	2	2	2	2	1	2	2	2	2	1	2	2
FSy	3	1	2	2	3	3	3	2	3	2	2	2	3	2	2	2	2	2
FD	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
FA	1	2	2	1	1	1	2	2	1	2	2	1	1	2	2	2	2	2
FB	2	2	2	2	2	2	2	1	1	1	2	1	1	1	1	1	2	2
FN	2	2	1	1	2	2	2	2	1	1	2	1	1	1	1	1	1	1
FPL	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
FDL	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
FR	2	1	2	2	2	2	1	2	2	1	1	1	1	1	2	1	1	1
Endoca	rps																	
ES	3	3	3	3	4	3	3	3	3	3	3	3	3	3	3	3	3	3
ESA	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
ESB	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
ED	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
EA	1	1	1	1	1	1	1	1	1	1	2	1	2	2	2	2	2	2
EB	3	3	3	3	2	2	2	2	2	3	3	3	3	3	3	3	3	3
ESu	2	2	2	2	2	2	2	2	2	2	2	2	2	1	2	2	2	2
ENG	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
EDG	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
EM	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2

BT1 to **BT20**: Olive trees from the BeniTajjit site; **EA**: Endocarp apex; **EB**: Endocarp base; **ED**: Position of maximum transverse diameter of endocarp; **EDG**: Distribution of the endocarp fibrovascular grooves; **EM**: Bill of the endocarp apex; **ENG**: Number of the endocarp fibrovascular grooves; **ES**: Endocarp shape; **ESA**: Endocarp symmetry (position A); **ESB**: Endocarp symmetry (position B); **ESu**: Endocarp surface; **FA**: Fruit apex; **FB**: Fruit base; **FD**: Position of maximum transverse diameter; **FDL**: Dimension of lenticels; **FN**: Fruit nipples; **FPL**: Presence of lenticels; **FS**: Fruit shape; **FR**: Ripeness start; **FSy**: Fruit symmetry; **IL**: Inflorescence length; **IN**: Number of the inflorescence flowers; **LC**: Longitudinal curvature of the leaf blade; **LS**: Leaf shape.

Difference between all studied olive trees was ranged from one as minimum value (so be it 4.3%) to nine characters (so be it 39.1%) as maximum value (Table 3). In this case, for BT3 -BT5; BT6 - BT7 or BT19 - BT20 olive pairs, difference was respectively just about the fruit apex form (rounded or pointed) or the endocarp shape (elongated or elliptic) or the leaf shape (lanceolate or elliptic – lanceolate). For BT6 - BT16 or BT6 – BT18 olive pairs, difference was maximal. Nine characters were implicated. In the first pair (BT6 - BT16), differences interest five fruit (symmetry, apex, base, nipple, and ripeness) and four endocarp (shape, apex, base, and surface) characters. In the second pair (BT6 – BT18), differences interest six fruit (shape, base, symmetry, apex, nipple, and ripeness) and three endocarp (shape, apex, and base) characters.

Table 3. Morphological character number discriminating studied olives one to one.

	BT1	BT2	BT3	BT5	BT6	BT7	BT8	BT10	BT11	BT12	BT13	BT14	BT1	BT1	BT1	BT18	BT19	BT20
						-		0	-	2	ω	4	15	16	17	∞	9	0
BT1	00																	
BT2	03	00																
BT3	03	03	00															
BT5	02	04	01	00														
BT6	02	05	05	04	00													
BT7	01	04	04	03	01	00												
BT8	03	02	04	05	03	02	00											
BT10	04	04	03	04	04	03	03	00										
BT11	03	06	04	03	03	02	04	03	00									
BT12	05	03	02	03	07	06	04	03	04	00								
BT13	05	03	04	05	07	06	04	05	08	04	00							
BT14	06	06	05	04	08	07	07	06	05	03	07	00						
BT15	05	06	06	05	07	06	06	07	04	04	06	03	00					
BT16	07	05	04	05	09	08	06	05	06	02	04	05	04	00				
BT17	05	05	02	03	07	06	06	03	04	02	04	05	04	02	00			
BT18	07	05	04	05	09	08	06	05	06	02	02	05	04	02	02	00		
BT19	06	04	03	04	08	07	05	06	07	03	03	04	03	03	03	03	00	
BT20	05	03	02	03	07	06	04	05	06	02	02	05	04	02	02	02	01	00
$BT \cdot O1$	ive tre	pes fro	m the	ReniT	Caiiit s	ite												

BT: Olive trees from the BeniTajjit site.

Hierarchical Cluster Analysis

Hierarchical clustering based on squared Euclidean distance calculated from the twenty three morphological character categories revealed the existence of five major groups of olive trees according to their aggregation distance values (Figure 1, Table 4). The first group included olives trees that had the lowest value of aggregation distance which is equal to number 1 (Table 4). In this group, olives trees were different for one to two morphological characters. This group could be subdivided in one's turn into three subgroups due to the type of organ which constitutes the difference. The second one included olive trees for which the aggregation distance value is about 1.35 (Table 4). This group is composed by the seven olive trees BT8, BT11, BT12, BT16, BT17, BT18, and BT13 in which the difference from is two to six morphological characters among those of fruit and endocarp. The third group contained olive trees for which aggregation distance value is about 1.7 (Table 4). This group is composed by the three olive trees BT2, BT10, and BT14 in which the difference is from four to six morphological characters concerning in the same time fruit, endocarp, leaf, and inflorescence. The fourth group formed by the only olive tree BT15 for which aggregation distance value is about 5.9 (Table 4). This olive tree presents three to seven morphological characters in difference with the other olive trees included in all previous groups. It is the most differentiated olive tree.

Morphological characteristics of the most known Moroccan olive varieties

The six Moroccan olive varieties that were used in this study were *Picholine marocaine*, *Menara*, *Haouzia*, *Meslala*, *Dahbia*, and *Bouchouika*. For their morphological characteristics, we used results presented previously by Idrissi and Ouazzani (2004) (Table 5). These results showed that these varieties would be distinguished among themselves for two as minimum (2/23 so be it 8.7%) to seventeen morphological characters as maximum (17/23 so be it 73.9%) (Table 6).

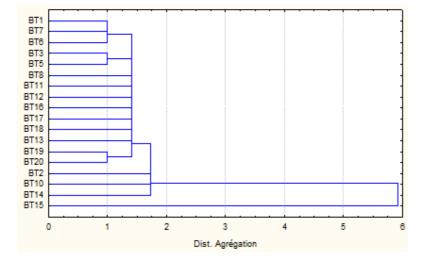


Figure Dendrogram 1. relationships showing the between the eighteen studied olive trees. Hierarchical clustering is based on squared Euclidean distance calculated from the twenty three morphological character categories. Accessions are named from **BT1** to **BT20**.

Table 4. Quantitative variables corresponding to the morphological characteristic categories of the eighteen olive trees which grouped by their aggregation distances that were determined previously by the hierarchic classification (UPGMA method). Characters that are identical in all olive trees are not presented here.

		First			ond	Th	ird									rd gro = 1.7		Forth group (d=5.9)
	st	ıbgro	up	-	roup		group											
	BT1	BT7	BT6	BT3	BT5	BT19	BT20	BT8	BT11	BT12	BT16	BT17	BT18	BT13	BT2	BT10	BT14	BT15
Leave	Leaves																	
LS	2	2	2	2	2	3	2	2	2	2	2	2	2	2	2	2	3	3
Inflor	esce	nces																
IL	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1	2	1
Fruits																		
FS	2	2	2	2	2	2	2	2	2	2	2	2	1	1	2	2	2	2
FSy	3	3	3	2	2	2	2	3	3	2	2	2	2	2	1	2	2	3
FA	1	1	1	2	1	2	2	2	1	2	2	2	2	2	2	2	1	1
FB	2	2	2	2	2	2	2	2	1	1	1	1	1	2	2	1	1	1
FN	2	2	2	1	1	1	1	2	1	1	1	1	1	2	2	2	1	1
FR	2	2	2	2	2	1	1	1	2	1	1	2	1	1	1	2	1	1
Endo	carp	S																
ES	3	3	4	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3
EA	1	1	1	1	1	2	2	1	1	1	2	2	2	2	1	1	1	2
EB	3	2	2	3	3	3	3	2	2	3	3	3	3	3	3	2	3	3
ESu PT: OI	2	2	2	2	2	2	2	2	2	2	1	2	2	2	2	2	2	2

BT: Olive tree from the Beni Tajjit site; **EA**: Endocarp apex; **EB**: Endocarp base; **ES**: Endocarp shape; **ESu**: Endocarp surface; **FA**: Fruit apex; **FB**: Fruit base; **FN**: Fruit nipples; **FS**: Fruit shape; **FR**: Ripeness start; **FSy**: Fruit symmetry; **IL**: Inflorescence length; **LS**: Leaf shape.

Comparison of the BT olive trees with the most known Moroccan olive varieties

The hierarchical cluster analysis based on the morphological data of both the BT olive trees and the six Moroccan olive varieties (Figure 2) and their comparison one to one by their morphological characters (Table 7) showed that:

- The BT14 olive tree is the nearest one to the *Picholine marocaine* variety. Aggregation distance value equal to 1,42 is the lowest one. Difference between these two olive cultivars interest two morphological characters (Lanceolate or elliptic-lanceolate leaves for BT14 and *Picholine marocaine* respectively and

Table 5. Quantitative variables corresponding to the morphological characteristic categories of the six most known Moroccan olive varieties (Data from Idrissi & Ouazzani, 2004).

	Picholine marocaine	Menara	Haouzia	Dahbia	Meslala	Bouchouika
Leaves						
LS	2 2	2 2	2 2	3 3	2 2	3 3
LS LC		2	2	3	2	3
Inflorescence						
IL IN	2 1	2 1	2 1	1 1	2 2	1 1
IN	1	1	1	1	2	1
Fruits						
FS	2	2	2	3	2	3
FSy	3	2	3	3	2	3
FD	2	2	2	2	2	2
FSy FD FA	2 3 2 1 1 1 2 1	2 2 2 1 1 2 1	2 3 2 1 1 1 2 2 3	3 3 2 2 2 2 1 1	2 2 2 2 2 2 2 2 2 2 2 2 2 2	3 3 2 2 2 2 2 1 1 2
FB	1	1	1	2	2	2
FN	1	1	1	2	2	2
FPL	2	2	2	1	2	1
FPL FDL	1	1	2	1	2	1
FR	1	2	3	3	2	2
Endocarps						
ES ESA ESB	3	3	3	4	3	4
ESA	2	2	2	2	2	2
ESB	1	1	1	2	1	2
ED	2	2	2	3	2	3
EA	1	1	1	1	2	1
EA EB	3	3	3	2	3	2
ESu	2	2	2	1	2	1
ENG	2	2	2	2	1	1
EDG	3 2 1 2 1 3 2 2 1 2	3 2 1 2 1 3 2 2 1 2	3 2 1 2 1 3 2 2 1 2	4 2 3 1 2 1 2 1 2	3 2 1 2 2 3 2 1 2 2	4 2 3 1 2 1 1 2 2 2
EDG EM	2	2	2	2	2	2

EA: Endocarp apex; **EB**: Endocarp base; **ED**: Position of maximum transverse diameter of endocarp; **EDG**: Distribution of the endocarp fibrovascular grooves; **EM**: Bill of the endocarp apex; **ENG**: Number of the endocarp fibrovascular grooves; **ES**: Endocarp shape; **ESA**: Endocarp symmetry (position A); **ESB**: Endocarp symmetry (position B); **ESu**: Endocarp surface; **FA**: Fruit apex; **FB**: Fruit base; **FD**: Position of maximum transverse diameter; **FDL**: Dimension of lenticels; **FN**: Fruit nipples; **FPL**: Presence of lenticels; **FS**: Fruit shape; **FR**: Ripeness start; **FSy**: Fruit symmetry; **IL**: Inflorescence length; **IN**: Number of the inflorescence flowers; **LC**: Longitudinal curvature of the leaf blade; **LS**: Leaf shape. weakly asymmetrical or asymmetrical fruits for BT14 and *Picholine marocaine*

Table 6. Number of the morphological charactersamongthetwentythreestudiedonesdistinguishingthemostknownMoroccanolivevarietiesone.

	Picholine marocaine	Menara	Haouzia	Dahbia	Meslala	Bouchouika
Picholine	0					
marocaine						
Menara	3	0				
Houzia	2	4	0			
Dahbia	14	14	14	0		
Meslala	10	7	9	17	0	
Bouchouika	16	15	17	3	14	0

respectively). The other BT olive trees are at aggregation distance equal to 1.73 with regard to the *Picholine marocaine* variety. They are distinct of the *Picholine marocaine* variety by three to seven morphological characters.

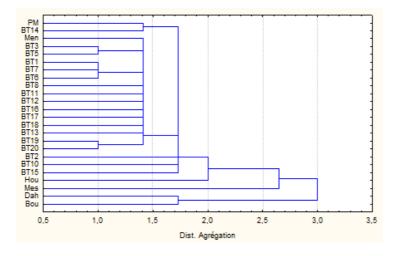
- The olive trees BT3, BT5, BT1, BT7, BT6, BT8, BT11, BT12, BT16, BT17, BT18, BT13, BT19, and BT20 are at aggregation distance of 1.42 with regard to the Menara variety. They are distinct of it by two to seven morphological characters. In this olive group, the BT3, BT12, and BT17 olive trees are the nearest ones to *Menara* variety. They have only two characters as difference.

- The olive trees BT2, BT10 and BT15 are at an aggregation distance of 1.73 with regard to either the *Picholine marocaine* or the *Menara* varieties. These olive trees are distinct to the *Picholine marocaine* variety by five, six and three morphological characters respectively and to the *Menara* variety by five, three and six morphological characters respectively.

- All BT olive trees are at an aggregation distance equal to:

(i) the value of 2 with regard to the *Haouzia* variety, with four to nine morphological characters as difference,

(ii) the value of 2.65 with regard to the *Meslala* variety, with seven to eleven morphological characters as difference,



Discussion

In Morocco, the number of distinct olive varieties is commonly referred as a single variety called 'Picholine marocaine' (Ouazzani et al., 1996; Khadari et al., 2007). Otherwise, this variety is usually cited as the most predominant variety considering that more than 98% of the olive growing orchards may be planted by it (Boulouha et al., 1992; Bamouh, 1998). However, at the beginning of the 19th century, some local olive cultivars were identified according to their morphological traits. Among them, for examples, were Bouchouk. Bouchouika, Fakhfoukha, Hamrani, Meslala, and Soussia (Maestratti, 1922; Torénzy, 1922). More tardily, some other varieties were then recorded: Dahbia, Haouzia, and Menara (Ouazzani et al., 1996). Haouzia and Menara, registered for cultivation in Morocco, were in fact developed through clonal selection (INRA, 2008). Menara is a direct selection of the Picholine marocaine variety; Haouzia a selection of both Picholine marocaine and Menara (FAO. 2009b). These new clones were then subsequently registered as Moroccan varieties (INRA, 2008). They show a large

(iii) the value of 3 with regard to the *Dahbia* and *Bouchouika* varieties, with nine to fourteen or with ten to sixteen morphological characters respectively as differences.

Figure 2. Dendrogram showing the relationships between studied Beni Tajjit olive trees and the six olive varieties which are the most known ones at the national scale. Hierarchical clustering is based on squared Euclidean distance calculated from the all morphological character categories studied. Accessions are named from BT1 to BT20. National olive varieties studied are Picholine marocaine (PM), (Men), Haouzia (Haou), Menara Meslala (Mes), Dahbia (Dah), and Bouchouika (**Bou**).

morphological similarity with *Picholine marocaine* variety. Among the 23 analysed characters, *Haouzia* variety shows 21 characters and *Menara* variety 20 characters identical to those of *Picholine marocaine* variety.

Results obtained here showed that the olive trees from the BT site exhibited some morphological differences between themselves and in comparison with each Moroccan analysed variety. Differences inside the BT olive trees group concerned between one to nine characters and with regard to the six Moroccan varieties they seem be ranged between two to sixteen characters. The BT14 olive tree with only 2 morphological characters (leaf shape and fruit symmetry) which differentiate it with the Picholine marocaine variety may be the nearest olive tree to this variety. It possessed. equally, the same morphological characters number (tow) as difference towards the *Picholine* marocaine variety than possessed Haouzia cultivar towards it. However, character categories concerned by this difference were not the same. Otherwise, the BT14 olive tree showed three (leaf shape, fruit

Table 7. Number of morphological characters showing differences between the most known Moroccan olive varieties on the one hand and between these varieties and the studied Beni Tajjit (BT) olive trees on the other hand.

Ch			Moroccan oliv			
Character numbers in difference	Picholine marocaine	Menara	Haouzia	Meslala	Dahbia	Bouchouika
0	PM	Men	Haou	Mes	Dah	Bou
1						
2	Haou, BT14	BT3, BT12, BT17	PM			
3	Men, BT11, BT12, BT15	PM, BT5, BT10, BT14			Bou	Dah
4	BT1, BT5	Haou, BT11, BT16, BT18, BT20	Men, BT11, BT14			
5	BT2,BT3, BT7, BT8, BT16, BT17, BT18, BT20	BT1, BT2, BT19	BT1, BT5, BT12, BT15			
6	BT6, BT10, BT19	BT7, BT8, BT13, BT15	BT3, BT7, BT17			
7	BT13	Mes, BT6	BT2, BT6, BT8, BT10, BT16, BT18, BT20	Men, BT3, BT13, BT17, BT20		
8			BT19	BT1, BT2, BT5, BT10, BT19		
9			Mes, BT13	Haou, BT7, BT8, B12, BT16, BT18	BT6, BT8	
10	Mes			PM, BT6, BT14	BT7	BT6
11				BT11, BT15	BT1, BT2, BT10	BT7, BT8
12					BT3, BT11, BT12, BT19	BT1, BT10
13					BT5, BT14, BT15, BT20	BT2, BT3, BT11
14	Dah	Dah	Dah	Bou	PM, Men, Haou, BT13, BT16, BT17, BT18	Mes, BT5, BT13, BT19
15		Bou				Men, BT12, BT15, BT16, BT17, BT20 PM, BT14,
16	Bou			•		BT18
17			Bou	Dah	Mes	Haou

PM: Picholine marocaine; Men: Menara; Haou: Haouzia; Mes: Meslala; Dah: Dahbia; Bou: Bouchouika.

apex, and fruit ripeness) and four (leaf shape, fruit symmetry, lenticels dimension, and fruit ripeness) different morphological characters towards *Menara* or *Haouzia* cultivars respectively. BT14 olive tree could be then the closer one to the *Picholine marocaine* variety and less to *Menara* and *Haouzia* cultivars. BT11, BT12, and BT15 olive trees resembled too greatly to the *Picholine marocaine* variety. Each one of these olive trees showed only three different morphological characters with this variety. BT3, BT12 and BT17 olive trees could be the nearest ones towards *Menara* cultivar. Each one showed two different characters with it. BT11 and BT14 olive trees with four different morphological characters seemed the closer ones to the *Haouzia* cultivar.

For the rest of the national olive cultivars *Meslala*, *Dahbia*, and *Bouchouika*, all BT olive trees were

40

morphologically very different with them. In fact, differences concerned between seven to sixteen characters among the twenty three analysed. It was reported that these three cultivars were cultivated in Morocco in restricted areas that are geographically so farther to our study site: Meslala and Dahbia near Meknès and Bouchouika near Sefrou (in Ouazzani et al.. 1996). Although some olive characteristics like leaf shape. inflorescence length, flower number, fruit shape, endocarp shape, etc. could vary due exogenous factors (environment, to cultivation technology, etc.) (Barronco & Rallo 1984; Cantini et al., 1999; Idrissi & Ouazzani, 2004), quantity and quality of morphological characters concerned by differences cannot explain large differences revealed between BT olive trees themselves and between BT olive trees and the six national olive cultivars. Resemblance may be high only in few cases like the BT14 olive tree and the Picholine marocaine variety.

Otherwise, if we consider the old ages of the BT olive trees (BT1 more than 80 years old; BT2 and BT3 near than 35 years old; BT5-BT11 near than 60 years old; BT12-BT20 near than 23 years old), we can easily remark that all BT olive trees were planted before the selection date of *Menara* and *Haouzia* clones and *a fortiori* before their multiplication and distribution in great scale. Indeed, in Beni Tajjit site, production of olive trees is still largely based on the traditional and local methods.

Conclusion

In this study, obtained results were a proof of variable heterogeneity in the morphological characteristics between the studied olive trees cultivated in the Beni Tajjit site. They present equally some differences with regard to the six most known olives varieties at the national Picholine marocaine. scale: Menara. Haouzia. Meslala, Dahbia, and Bouchouika. This confirms previous results that conclude that varietal diversity in

This present work showed that our study site could contain new olive varieties other than those previously known at the national scale. This diversity could be more enlarged in seen that this study was conducted on a small olive trees sample and on a limited area of BT site where the superficies cultivated by olives reach 760 ha. These occasional records suggest that varietal diversity in Moroccan olive growing orchards may be much higher than what was assessed previously. This idea is in agreement with that formed previously by Ouazzani et al. (1996). In the same way, results obtained by other authors recorded genetic and phenotypic heterogeneities in the Moroccan olive orchard (Ouazzani et al., 1996; Ouazzani et al. 1997; Lumaret et al., 2000; Essadki & Ouazzani, 2003, Idrissi & Ouazzani, 2004). Discrimination of varieties based on morphology evaluation like that we have made is limited by its requirement to a well-trained staff and influenced by cultural and environmental conditions (Belaj *et al.*, 2001). To a better characterization of the olive cultivars, different molecular techniques should be used (Rallo et al., 2000; Bandlej et al., 2002). They had been successfully applied diversity analysis for of varietal identification and characterization (Rekik et al., 2008; Muzzalupo et al., 2009, 2010) and to address the issue of olive homonymy and synonymy (Khadari et al., 2003; Bracci et al., 2009).

Moroccan olive material may be much higher than what was assessed previously. In the future, it is important to extent the research with more locally as well as regionally investigations. Studies of genetic resources could be particularly important for preserving the biodiversity and maintaining the advantages of local cultivars which are well adapted to local conditions. Morphological study could be completed by molecular methods which are very suitable to reach a better understanding of the material's genetic diversity. Both, morphological and molecular data could be compared in order

References

Baldoni L, Cultrera NGM, Mariotti R, Ricciolilni C, Arcion S, Vendramin GG, Bounamici A, Porceddu A, Sarri V, Ojeda MA, Trujillo I, Rallo L, Belaj A, Perri E, Salimonti A, Muzzalupo I, Casagrande A, Lain O, Messina R, Testolin R (2009) A consensus list of microsatellite markers for genotyping. Mol Breeding **24**: 1-19.

Bamouh A (1998) Plan National oléicole: Les axes d'intervention et le plan d'action 1998-2010. Bull. Transfert de Technologie **51**: 1-4.

Bandelj D, Jakše J, Javornik B (2002) DNA fingerprinting of olive varieties by microsatellite markers. Food Technol Biotech. **40**: 185-190.

Bandelj D, Jakse J, Javornik B (2004) Assessment of genetic variability of olive variety by microsatellite and AFLP markers. Euphytica **136**: 93-102.

Barranco D, Rallo L (1986) Gordalsevillana. Olivae **13**: 34-35.

Barranco D, Rallo L (1984) Las variedades de olivocultivadas en Andalucia. Edition M. de Agricultura, Junta de Andalucia, Madrid, Spain.

Barranco D, Cimato A, Fiorino P, Rallo L, Touzani A, Castaneda C, Serafin F, Trujillo I (2000) World catalogue of olive varieties. Edition Internacional Olive Oil Council, Madrid, España.

Belaj A, Trujillo I, Rossa R, Rallo L, Gimenez MJ (2001) Polymorphism and discrimination capacity of randomly amplified polymorphic markers in an olive germplasm bank. J. Amer. Soc. Hort. Sci. **126**: 64-71.

Besnard G, Batadat P, Chevalier D, Tagmount A, Berville A (2001) Genetic differentiation in the olive complex (*Olea europaea*) revealed by RAPDs and RFLPs in the rRNA genes. Genet. Resources Crop. Evol. **48**: 165-182. to detect the level of reliability for the morphological parameters and to provide information on which parameters should be useful to discriminate olive cultivars.

Boulouha B, Loussert R, Saadi R (1992) Etude de la variabilité phénotypique de la variété 'Picholine marocaine' dans la région du Haouz. Olivae **43**: 30-33.

Bracci T, Sebastiani L, Busconi M, Fogher C (2009) SSR markers reveal the uniqueness of olive cultivars from the Italian region of Liguria. Sci. Hort. **122**: 209-215.

Cantini C, Cimato A, Graziano S (1999) Morphological evaluation of olive germplasm present in Tuscany region. Euphytica **109**: 173-181.

COI (1997)Méthodologie de caractérisation primaire des variétés d'olivier. Projet sur la conservation, caractérisation, collecte et utilisation de génétiques ressources de l'Olivier. (RESGEN-T96/97). Edition Union Européenne, Conseil Oléicole International, Madrid, España.

Erfatpour M, Hamidogli Y, Kaviani B, Fatahi R, Falahati M, Javadi D, Hashemabadi D (2011) Assessment of genetic diversity among some Iranian hazelnut genotypes using SSR markers. Aust. J. Crop Sci. **5**: 1286-1291.

Essadki M, Ouazzani N (2003) Résultats préliminaires de l'identification des variétés d'olivier à l'aide des marqueurs génétiques ISSR. Olivae **97**: 42-45.

FAO (2009b) Olive Gap Manual. Good Agricultural Practices For the near East & North Africa Countries. Food and Agriculture Organization of the United Nations regional office for the near East. Edition FAO, Cairo.

FAO (2009a) Agricultural Statistics of the Food and Agriculture Organization of the United Nations. Edition FAO. Rome.

FAOSTAT (2006) FAO statistical databases. Agriculture data collection (primary crops). http://apps.fao.org/page/

collections?subset=agriculture [accessed on June 14, 2008].

Gomes S, Martins-Lopes P, Lima-Brito J, Meirinhos J, Lopes J, Martins A, Guedes-Pinto H (2008) Evidence for clonal variation in 'Verdeal-Transmontana' olive using RAPD, ISSR and SSR markers. J Hortic. Sci. Biotech. **83**: 395-400.

Idrissi A, Ouazzani N (2004) Apport des descripteurs morphologiques à l'inventaire et à l'identification des variétés d'olivier (*Olea europaea* L.). Plant Genet. Resour. Newslett. **136** : 1-10.

INRA (2008) L'Olivier: Sélection de variétés en irrigué. Rapport d'activité, INRA 2007, Rabat, Morocco: 22-23.

Khadari B, Charafi J, Moukhli A, Ater MA (2007) Despite a single major cultivar: a paradoxical situation evidenced by the use of SSR loci. Tree Genet Genome **4**: 213-221.

Khadari B., Breton C, Moutier N, Roger JP, Besnard G, Berville A, Dosba F (2003) The use of molecular markers for germplasm management in a French olive collection. Theor. Appl. Genet. **106**: 521-529.

La Mantia M., Lain O, Caruso T, Testolin R (2005) SSR-based DNA fingerprints reveal the genetic diversity of Sicilian olive (*Olea europaea* L.) germplasm. J. Hort. Sci. Biotechnol. **80**: 628-632.

Lumaret R, Amane M, Ouazzani N, Baldoni L (2000) Chloroplast DNA variation in the cultivated wild olive taxa of the genus *Olea* L. Theor. Appl. Genet. **101**: 547-553.

Maestratti I (1922) Pour l'olivier. In : Maurice M (ed.) Proceedings of the Fifth International olive Congress, Marrakech, Morocco, September 26 to October 10, 1922.

Ministère de l'Agriculture et de la Pêche Maritime, Maroc - Direction de la Stratégie et des Statistiques, Note stratégique n°95, Veille économique-Secteur oléicole, Septembre 2013. Montemurro C, Simeone R, Pasqualone A, Ferrara E, Blanco A (2005). Genetic relationships and cultivar identification among 112 olive accessions using AFLP and SSR markers. J. Hort. Sci. Biotechnol. **80**: 105-110.

Msallem M, Mehri H, Radhouane L (2000) Inventaire des collections d'olivier en Tunisie. Plant Genet. Ressour. Newslett. **122**: 36-40.

Muzzalupo I, Chiappeta A, Benincasa C, Perri E (2010) Intra-cultivar variability of three major olive cultivars grown in different areas of central-southern Italy and studied using microsatellite markers. Sci. Hortic. Amsterdam **324**: 324-329.

Muzzalupo I, Stefanizzi F, Salimonti A, Falabella R, Perri E (2009) Microsatellite markers for identification of a group of Italian olive accessions. Sci. Agric. (Piracicaba, Braz) **66**: 685-690.

Ouazzani N, Lumaret R, Villemur P (1996) Genetic variation in the olive tree (*Olea europaea* L.) cultivated in Morocco. Euphytica **91**: 9-20.

Ouazzani N, Lumaret R, Villemur P, Amane M (1997) Ressources génétiques de l'Olivier (*Olea europaeaL.*) au Maroc. *Proceedings Séminaire International « Ressources phytogénétiques et développement durable»*, Actes éditions, Octobre 1994, Rabat, Maroc : 313-318.

Ouazzani N, Lumaret R, Villemur P, Di Giusto F (1993) Leaf allozyme variation in cultivated and wild olive trees (*Olea europaea* L.). J. Hered. **84**: 34-42.

Poljuha D, Sladonja B, BrkicBubolo K, Radulovic M, Brscic K, Setic E, Krapac M, Milotic A, (2008) A multidisciplinary approach to the characterisation of autochthonous Istrian olive (*Olea europeaL.*) varieties. Food Technol. Biotech. **46**: 347-354.

Rallo P, Dorado G, Martin A (2000) Development of simple sequence repeats (SSRs) in olive tree (*Olea europaea* L.). Theor. Appl. Genet. **101**: 984-989. Rekik I, Salimonti A, Grati-Kamoun N, Muzzalupo I, Perri E, Rebai A, (2008) Characterisation and identification of Tunisian olive tree varieties by microsatellite markers. Hort. Science **43**:1371-1376.

Ruby J (1916) Recherches morphologique et biologique sur l'olivier et sur ses variétés cultivées en France. Ann. Sci. Nat. Bot. **20**: 1-286.

Rugini E, De Pace C, Gutierrez-Pesce P, Muleo R (2011). *Olea*. In: Wild Crop Relatives: Genomic and Breeding Resources, Temperate Fruits. Kole C, Edition Springer-Verlag Berlin Heidelberg, Berlin.

Shabanimofrad M, Yusop MR, Saad MS, Wahab PEM, Biabanikhanehkahdani A, Latif MA (2011) Diversity of physic nut (*Jatropha curcas*) in Malaysia: application of DIVA-geographic information system and cluster analysis. Aust. J. Crop. Sci. 5: 361-368. Therios I (2005) Olive Production. Gartaganis Publications, Thessaloniki, Greece: 476 pp.

Tornezy M (1922) L'oléiculture dans la région de Marrakech. Maurice M, Edition Proceedings of the Fifth International olive Congress, Marrakech, Maroc, September 26 to October 10, 1922.

Tous J, Romero A (1992) Arbequina. Olivae **43**: 28-29.

Trigui A, Msellem M (2002) Olivier de Tunisie, Catalogue des variétés autochtones et types locaux, VI, IRESA, Institut de l'olivier, Tunisie.

Trujillo I, Rallo L, Arus P (1995). Identifying olive cultivars by isozyme analysis. J. Amer. Soc. Hort. Sci. **120**: 318-324.

Ziliotto F, Barcaccia G, Baldoni L, Tonutti P (2002) Identificazione e caratterizzazione di alcune cultivar di olivo. L'informatore agrario **15**: 115-118.