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# Detection of genetic diversity in Moroccan durum wheat accessions using agro-morphological traits and microsatellite markers

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Characterization of germplasm by agro-morphological, biochemical and DNA-markers provide powerful tools to precise germplasm identification, quantify the genetic diversity and to estimate the phylogenetic relationship among accessions in many crop species. The objective of the present study was to characterize a subset of 23 Moroccan relevant durum wheat (Triticum turgidum L. var. durum) accessions selected from the National Gene Bank of Morocco for their genetic diversity using agromorphological and microsatellite markers, and to measure the genetic distance among these accessions using UPGMA cluster analysis. Durum wheat accessions were planted under field condition and their agro-morphological characters such as days to emergence, days to tillering, days to booting, days to head emergence, days to flowering, days to physiological maturity, plant height, and thousands kernel weight were recorded. The same set of accessions was also analyzed for polymorphism with 7 genomic microsatellite markers. The results indicated that the evaluated germplasm comprises of useful variation for future breeding activities. Furthermore, the cluster analysis based on microsatellite data showed the closest correlation with the groupings of particular genotypes based on agro-morphological characters. The authors results suggest that the characterization based on agro-morphological traits and genotypic markers will be a useful tool to the breeders to choose genotypes with appropriate diversity.

Key words: Durum wheat, *Triticum* germplasm, agro-morphological and microsatellite markers, genetic diversity.

# INTRODUCTION

Durum wheat (*Triticum turgidum* L.var. *durum*) is an important species of the tribe Triticeae, and is one of the most important food crops in the world because of its adaptation to semi-arid environments and its unique end products. It is an allotetraploid (genome AABB) with a basic chromosome number of x = 7 and 2n = 4x = 28. In

the West Asia and North Africa (WANA) region, and in the Mediterranean basin, durum wheat has historically received special attention as a major crop (Belaid, 2000). In Morocco durum wheat is an economically and nutritionally important cereal crop. It is grown in an area of 1.1 to 1.3 million hectares annually, ranks third after barley and bread wheat (MADRPM, 2008). Also, Morocco has been considered as a center of genetic diversity for *Triticum* genera (Vavilov, 1926). According to Fufa et al. (2005), knowledge of genetic diversity of elite breeding materials has been successfully used for efficient germplasm management, genotype selection for different plant breeding purposes, and the conservation of genetic resources. Therefore, precise identification and

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Abbreviations: bp, Base pairs; SSR, simple sequence repeat or microsatellite; UPGMA, unweighted pair group method of arithmetic average.

characterization of the conserved accessions is of great value for quantifying the extent of intraspecific genetic diversity within accessions, detecting duplications of genetic material in germplasm collection, improving and securing durum wheat culture in Morocco and in the world.

The conventional methods used to characterize and identify accessions in wheat species are based on morphological characterization and phenological observations according to UPOV (Unité pour la Protection des Obtentions Végétales) and IPGRI (now, known as Bioversity International) descriptors. Morphological traits have been studied for the estimation of genetic diversity and as selection criteria for wheat breeding (Schut et al., 1997; Marić et al., 1998; Casadesus et al., 2007; Marti et al. 2007). The disadvantages of this conventional approach of characterization are the high cost, timeconsuming, low polymorphism and heritability, and the influences of environmental factors. Also morphological character are often limited in their numbers and may be controlled by epistatic and pleitropic gene effects (Van Beuningen and Busch, 1997). Thus these characters may not adequately represent the genetic diversity among genotypes.

Conversely, identified genetic variations in plant germplasm based on DNA polymorphism are abundant and independent of environmental factors. Furthermore, a relatively small sample size can be informative for the evaluation of genotypes when DNA polymorphisms are analyzed. In contrast a large sample size is required when quantitative traits are measured. Therefore, characterization at the DNA level may be much less timeconsuming and less labor intensive. DNA markers that differentiate genotypes are more reliable and convenient than physiological or morphological characters in the identification and characterization of genetic variation (Rao and Riley, 1994; Weising et al., 1995; Gupta et al., 1999; Prasad et al., 2000; Almanza-Pinzón et al., 2003; Naghavi et al., 2004, 2007; Pagnotta et al., 2005; Udupa et al., 1999).

Triticum genera have been used for a number of cvtogenetic and taxonomic studies (Gupta and Shephered. 1990; Barkworth, 1992; Heslop-Harrison, 1992) but more recently, new molecular methods have been used for the study of genetic diversity and interspecific/intergeneric relationships among a number of species of the tribe Triticeae and among different species of Triticum. Among different types of molecular markers, there is a class of sequences, termed microsatellites (Litt and Luty, 1989), also known as simple sequence repeats (SSRs) DNA markers or simple tandem repeats (STRs) (Tautz et al., 1986). They are defined as tandemly repeated DNA sequences of 1 - 6 base pairs (bp) in length, characterized by their repeated motif and their number of repeats. They are codominant markers of relatively small size, which can be easily amplified with the polymerase chain reaction. Their simplicity, high level of polymorphism, high reproducibility makes them popular

for evolutionary and genetic diversity studies. Genomic microsatellites have been used in wheat for genome mapping, physical mapping, gene tagging, and genetic diversity estimates (Röder et al., 1998; Prasad et al., 2000; McLauchlan et al., 2001). Here, we characterize the genetic diversity within a subset of Moroccan durum wheat germplasm using agro-morphological characters and microsatellite markers and study the relationship between genotypic and phenotypic variation.

#### MATERIALS AND METHODS

#### Plant material

For this study a set of 23 accessions of durum wheat (*Triticum turgidum* L.var. *durum*) was used, along with 4 cultivars included as checks: 'Kyperounda' (2777), 'Tomouh', 'Marzak' and 'Acsad 65'. The details of these taxa are presented in Table 1. All the durum wheat accessions analyzed in this work was obtained from the National Gene Bank of Morocco, Centre Régional de la Recherche Agronomique de Settat, INRA, Settat, Morocco.

#### Measurements of agro-morphological characters

The trial was conducted at INRA Research Experimental Station at Sidi Al Aydi (31° 15' N, 7° 30' W), near Settat, Morocco. The soil is a vertic calcixeroll and has a depth of 90 to 120 cm. A collection of durum wheat accessions and 4 checks namely 'Kyperounda', 'Marzak', 'Tomouh', and 'Acsad 65' were used in the experiment. These four checks were augmented with 16 randomly allocated new entries (so, the block had 20 plots). The design followed an augmented block design. Each entry was sown in two rows of one meter long and 20 cm apart. The data reported in this article is for a subset of 19 accessions and four checks; in which molecular work has been undertaken (agro-morphological data for the sample no. 14 was not available due to unavoidable reasons). During the investigation eight quantitative agro-morphological traits namely days to emergence (EME), days to tillering (TIL), days to booting (BOO), days to head emergence (HEM), days to flowering (FLO), days to physiological maturity (MAT), plant height (HGT), and thousands kernel weight (TKW) were recorded. The phenological data were recorded according to the scale described by Zadoks et al. (1974).

#### Agro-morphological evaluations

The best use of the information contained in the data for agromorphological characterization is an important issue in plant breeding. To display the genetic variability among durum wheat genotypes, a Genotype x Trait biplot (GT biplot) of standardized data was applied. To generate a GT biplot (Yan et al., 2000), the genotype  $\times$  trait two-way table of data was first trait-standardized. The standardization is necessary to remove the units, because different traits use different units. The trait-standardized table (data standardized) was then decomposed into principal components (PC). The first two PC (PC1 and PC2) were used to generate a GT biplot. PC1 and PC2 were scaled so that values are symmetrically distributed between the genotype scores and trait scores. A genotype by trait biplot is constructed by plotting the PC 1 scores against the PC 2 scores for each genotype and each trait. The biplot technique (Gabriel, 1971, 1981; Gabriel and Odoroff, 1990; Kempton, 1984) provides a powerful tool for data analysis of genotype x trait data in individual environments and can be used to visualize the genetic correlations among traits and evaluation of the

Accession code	Accession number	Origin (province)							
1	570	Morocco (Kalaa Sraghna)							
2	682	Morocco (Azilal)							
3	483	Morocco							
4	656	Morocco							
5	'Képyrounda'	Morocco							
6	47	Unknown							
7	1	Morocco							
8	522	Morocco							
9	606	Morocco (Azilal)							
10	'Tomouh'	Morocco							
11	699	Morocco (Azilal)							
12	388	Morocco							
13	612	Morocco							
14	-	Morocco(Kalaa Sraghna)							
15	359	Morocco (Marrakech)							
16	177	Morocco							
17	313	Morocco (Khenifra)							
18	'Acsad 65'	Morocco							
19	'Marzak'	Morocco							
20	686	Morocco (Azilal)							
21	663	Morocco (Taza)							
22	335	Morocco (Tetouan)							
23	575	Morocco (Figuig)							

 Table 1. Accession code, accession number, and origin of 23 durum wheat accessions used in the experiment.

Table 2. Microsatellite markers used for diversity analysis of 23 durum wheat accessions used in the experiment.

Locus	Chromosomal location	Left primer (5'→3')	Right primer (5'→3')	Repeat motif in Chinese Spring	Annealing temperature used for PCR (°C)			
Xgwm132	6B	TACCAAATCGAAACA CATCAGG	CATATCAAGGTCT CCTTCCCC	(GA) <sub>24</sub> (GAA) <sub>6</sub> imp	55			
Xgwm146	7B	CCAAAAAAACTGCCT GCATG	CTCTGGCATTGCT CCTTGG	(GA)5GC(GA) <sub>20</sub>	55			
Xgwm160	4A	TTCAATTCAGTCTTG GCTTGG	CTGCAGGAAAAAA AGTACACCC	(GA) <sub>21</sub>	55			
Xgwm313	4A	GCAGTCTAATTATCT GCTGGCG	GGGTCCTTGTCTA CTCATGTCT	?	55			
Xgwm369	3A	CTGCAGGCCATGATG ATG	ACCGTGGGTGTTG TGAGC	(CT) <sub>11</sub> (T) <sub>2</sub> (CT) <sub>21</sub>	55			
Xgwm389	3B	ATCATGTCGATCTCC TTGACG	TGCCATGCACATT AGCAGAT	(CT) <sub>14</sub> (GT) <sub>16</sub>	55			
Xgwm493	3B	TTCCCATAACTA AAACCGCG	GGAACATCATTTC TGGACTTTG	(CA) <sub>43</sub> imp	55			

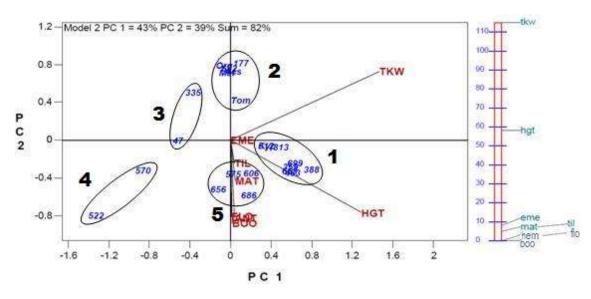
genotype on the basis of multiple traits (Yan, 2001; Yan et al., 2000, 2001; Yan and Rajcan, 2002; Yan and Kang, 2003; Lee and Petersen, 2002, 2003; Yan and Tinker, 2006). The GGE biplot software (Yan, 2001) was used for all calculations.

#### DNA extraction and microsatellite marker analysis

Total genomic DNA was extracted for each accession from

approximately 0.1 g of fresh young leaf collected from a 2 weeks old plant (one plant/accession). DNA was isolated using a cethyl trimetrhylammonium bromide (CTAB) method adapted from Udupa et al. (1998). The seven loci located on A and B genomes (Table 2) were chosen randomly from the set of microsatellites developed by Röder et al. (1998) in bread wheat, *Triticum aestivum* (L.) Thell. (2n =  $6 \times = 42$ ).

Amplification of genomic DNA was done according to Udupa et al. (1999) in a PCR reactions 10  $\mu l$  containing 50 ng of template DNA, 1 x



**Figure 1.** The durum wheat genotype by trait biplot based on 23 genotypes x 8 agromorphological traits two-way table. The numbers indicated in blue in figure denotes accessions number as indicated in Table 1. EME: days to emergence, TIL: days to tillering, BOO: days to booting, HEM: days to head emergence, FLO: days to flowering, MAT: days to physiological maturity, HGT: plant height and TKW: thousands kernel weight. PC1 and PC2 are the first and second principal components, respectively, accounting for 82% of total variance. Marked groups (1 - 5) are discussed in the text.

PCR Buffer, 0.2 mM dNTPs, 10 pmole of each primer and 1 unit of *Taq* polymerase. The amplification profile consisted of an initial period of DNA denaturation and *Taq* polymerase activation at  $94^{\circ}$ C for 2 min, followed by 25 cycles of  $94^{\circ}$ C for 5 s,  $55^{\circ}$ C for 10 s, and 72°C for 10 s. A final extension was done at 72°C for 6 minbefore cooling to 4°C. PCR products were resolved on a 6% denaturing polyacrylamide gel. After electrophoresis, the DNA bands were visualized by silver-staining.

#### Data analysis

The SSR markers were scored for the presence (1) or absence (0) of amplified bands of microsatellites for each of 23 samples. The microsatellite binary data matrix was used to calculate the genetic distance for each pair of accessions using the NTSYS-pc 2.0 program (Rholf, 1998). The genetic distance (shared index method) was used for cluster analyses (Unweighted Pair Group Method of Arithmetic Average; UPGMA) using NTSYS-pc 2.0 program and a dendrogram was generated.

## RESULTS

## Agro-morphological evaluations

To display the genetic variability among durum wheat genotypes, a principal component analysis of standardized data was applied to display durum wheat trait relationships, and its application in genotype characterization, and comparison. Because different traits use different units, the data standardization is necessary to remove the units. Principal components, PC 1 and PC 2 were scaled so that values are symmetrically distributed between the genotype scores and trait scores. A genotype by trait biplot is constructed by plotting the PC 1 scores against the PC 2 scores for each genotype (23) and each trait (8).

The genotype by trait biplot effectively reveals the interrelationships among durum wheat traits (Figure 1). It also provides a tool for visual comparison among genotypes based on multiple traits. The results of the genotype by trait biplot, explained 82% of the total variation, and are a good approximation of the total variation of the standardization data (Figure 1). Thousands kernel weight, plant height, days to booting, days to head emergence, days to flowering and days to maturity had relatively long vectors, suggesting that there was relatively large variation among accessions. In contrast, days to emergence and days to tillering had short vectors, suggesting that there was little or no variation among accessions.

The cosine of the angle between the vectors of two traits measures the similarity or the correlation between them relative to their variation among genotypes. Thus, an angle of zero indicates a correlation of +1, an angle <  $90^{\circ}$  suggests a positive correlation, an angle of  $90^{\circ}$  indicates no (0) correlation, implying independence, an angle >  $90^{\circ}$  indicates negative correlation, and an angle of  $180^{\circ}$  represents a correlation of -1. Thus, days to tillering, days to booting, days to head emergence, days to flowering and days to maturity and plant height had acute (<  $90^{\circ}$ ) angles between them, indicating that their variation were similar. On the contrary this phenological stages had obtuse (> $90^{\circ}$ ) angles with thousands kernel

weight, indicating that their variation were opposite to that of thousands kernel weight. Plant height had a near-right angle with thousands kernel weight, indicating that its variation was more or less independent of this trait. The genotypes studied could be grouped into 5 groups:

1. Group 1 contains mostly taller and late maturing genotypes. This group includes the tallest accession number 388 and the late maturing variety 'Kyperounda' (included as a check).

2. Group 2 contains the early maturing genotypes. This group includes the early maturing variety 'Acsad 65' (included as a check).

3. Group 3 contains 2 accessions number 47 and 335 which are mid-season maturing and shorter accessions, compared to Group 1 and 2.

4. Group 4 contains shorter, moderately late maturing, with lower thousands kernel weight (TKW) accessions. This group includes accession number 522, which is the shortest among all accessions.

5. Group 5 contains the late maturing accessions. This group includes the genotype number 656, which is the very late maturing accession among all the accessions.

Thousands kernel weight (TKW) depends on the cosine of the angle between the genotype and TKW vectors. Group 1 has a high value, in the opposite; group 4 has a low value.

## Microsatellite marker analysis and genetic distance

Seven microsatellite markers of bread wheat were used to test polymorphism between accessions of durum wheat. The microsatellites markers used are presented in Table 2. Three primers Xgwm146, Xgwm493 and Xqwm313 of the seven primers used generated a polymorphic pattern. The size of the amplified bands ranged from 50 to 150 bp. The highest number of polymorphic bands was observed with Xgwm146 locus located on chromosome 7B and at this locus 9 different alleles were observed. The second locus Xgwm493 located on chromosome 3B yielded 5 alleles and the third locus Xgwm313 (not mapped) yielded 3 alleles. The genetic distance (dissimilarity index) calculated based on microsatellite polymorphism varied greatly (Table 3). The highest genetic distance (0.7538) was observed between accessions 5 which was an old improved variety named 'Képyrounda' and accessions 2, 3, 10, 11, 12, 17, and 20 which were local landraces. The lowest genetic distance (0.0000) was observed between accessions 8 and 1, 9 and 21, and 11 and 20.

Based on the genetic distance, the cluster analysis was performed using UPGMA method (Figure 2) to study the relationships among the accessions. The analysis classified the entire accessions into 5 major groups namely, A, B, C, D and E. The dendrogram was rooted by an old cultivar 'Kyperounda'. Most of the resulting groups are strongly related by their phenological stages. Groups A and E contain the majority of tallest and late maturing genotypes which represents group 1; group B contains the majority of early maturing genotypes of the group 2; Group C contains accessions of group 3, which are mid-season maturing accession; and Group D contains accessions of group 4, which are short and moderately late, based on agro-morphological characteristics.

## DISCUSSION

The present study of genetic diversity is based on both the variation of agro-morphological traits and the polymorphism of microsatellite markers. The results showed that local durum accessions were variable for most of the evaluated agro-morphological traits and molecular markers. In the durum wheat, genotype by trait biplot (Figure 1), thousands kernel weight, plant height, days to booting, and days to flowering had long vectors. In other words they had large variation among genotypes studied, suggesting that they are the most discriminator of the quantitative agro-morphological data evaluated. Also, phenological stages had acute (< 90°) angles between them, indicating a positive correlation between them, particularly between days to booting and days to flowering, so days to booting can be recorded instead of the other phenological stages. On the contrary all phenological stages had obtuse (> 90°) angles with thousands kernel weight, indicating a negative correlation between them and that of thousands kernel weight. A near zero correlation was between thousands kernel weight and plant height as indicated by their near perpendicular vectors, suggesting that their variation was more or less independent.

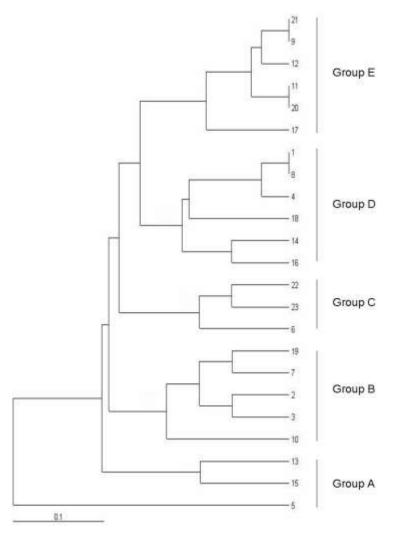
The twenty-three durum wheat genotypes evaluated using seven microsatellites revealed a high level of genetic variation. The numbers of alleles detected by the three microsatellites primers Xgwm146, Xgwm493 and Xgwm313 ranged from 3 to 9. The maximum number of alleles was observed at Xgwm146 locus located on The results indicate also that chromosome 7B. microsatellite loci of the B genome are more variable than those of the A genome. Cluster analysis of the durum accessions using molecular data shows some similarities and differences (Figure 2). The high genetic distance was observed between accessions 5, which is an old improved variety named 'Kyperounda' and the local landraces. This is a clear indication that the extent of diversity of genotypes selected from local germplasm was high and those accessions should be targeted for utilization in future genetic enhancement. Lack of microsatellite variation observed between some of the accessions could be due to genetic or failure to detect polymorphism in part because of less number of microsatellite markers used for the characterization or

Accessions number	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23
1	0	0.435	0.435	0.061	0.348	0.348	0.268	0	0.348	0.435	0.268	0.435	0.348	0.268	0.268	0.125	0.268	0.1942	0.125	0.268	0.348	0.531	0.348
2	0.435	0	0.125	0.531	0.754	0.348	0.125	0.435	0.530	0.268	0.435	0.636	0.194	0.268	0.435	0.435	0.435	0.348	0.268	0.435	0.531	0.348	0.531
3	0.435	0.125	0	0.531	0.753	0.348	0.125	0.435	0.530	0.268	0.435	0.636	0.348	0.268	0.636	0.435	0.435	0.348	0.268	0.435	0.531	0.194	0.348
4	0.061	0.531	0.531	0	0.435	0.435	0.348	0.060	0.268	0.531	0.194	0.348	0.435	0.348	0.348	0.194	0.194	0.268	0.194	0.194	0.268	0.636	0,435
5	0.348	0.7534	0.754	0.435	0	0.636	0.531	0.348	0.636	0.754	0.754	0.754	0.435	0.531	0.531	0.531	0.754	0.636	0.531	0.754	0.636	0.636	0.636
6	0.348	0.348	0.348	0.435	0.636	0	0.348	0.348	0.268	0.348	0.348	0.348	0.435	0.348	0.348	0.194	0.348	0.268	0.194	0.348	0.268	0.268	0.125
7	0.268	0.125	0.125	0.348	0.531	0.348	0	0.268	0.531	0.268	0.435	0.636	0.194	0.268	0.435	0.435	0.435	0.348	0.125	0.435	0.531	0.348	0.531
8	0	0.435	0.435	0.061	0.348	0.348	0.268	0	0.348	0.435	0.268	0.435	0.348	0.268	0.268	0.125	0.268	0.194	0.125	0.268	0.348	0.531	0.348
9	0.348	0.530	0.531	0.268	0.636	0.268	0.531	0.348	0	0.348	0.061	0.061	0.435	0.531	0.348	0.348	0.194	0.268	0.348	0.061	0	0.435	0.268
10	0.435	0.268	0.268	0.530	0.754	0.348	0.268	0.435	0.348	0	0.268	0.435	0.5301	0.435	0.636	0.435	0.435	0.348	0.268	0.268	0.348	0.531	0.531
11	0.268	0.435	0.435	0.194	0.754	0.348	0.435	0.268	0.061	0.268	0	0.125	0.531	0.435	0.435	0.268	0.125	0.194	0.268	0	0.061	0.531	0.348
12	0.435	0.636	0.636	0.348	0.754	0.348	0.636	0.435	0.061	0.435	0.125	0	0.531	0.636	0.435	0.435	0.268	0.348	0.435	0.125	0.061	0.531	0.348
13	0.348	0.194	0.348	0.435	0.435	0.435	0.194	0.348	0.435	0.531	0.531	0.531	0	0.348	0.194	0.531	0.531	0.435	0.348	0.531	0.435	0.268	0.435
14	0.268	0.268	0.268	0.348	0.531	0.348	0.268	0.268	0.531	0.435	0.435	0.636	0.348	0	0.636	0.125	0.435	0.348	0.435	0.435	0.531	0.194	0.348
15	0.268	0.435	0.636	0.348	0.531	0.348	0.435	0.268	0.348	0.636	0.435	0.435	0.194	0.636	0	0.435	0.435	0.194	0.268	0.435	0.348	0.531	0.348
16					0.531								0.531	0.125	0.435	0	0.268	0.194	0.268	0.268	0.348	0.348	0.194
17	0.268	0.435	0.435	0.194	0.754	0.348	0.435	0.268	0.194	0.435	0.125	0.268	0.531	0.435	0.435	0.268	0	0.194	0.268	0.125	0.194	0.531	0.348
18	0.194	0.348	0.348	0.268	0.636	0.268	0.348	0.194	0.268	0.348	0.194	0.348	0.435	0.348	0.194	0.194	0.194	0	0.194	0.194	0.268	0.435	0.268
19	0.125	0.268	0.268	0.194	0.531	0.194	0.125	0.125	0.348	0.268	0.268	0.435	0.348	0.435	0.268	0.268	0.268	0.194	0	0.268	0.348	0.531	0.348
20	0.268	0.435	0.435	0.194	0.753	0.348	0.435	0.268	0.061	0.268	0	0.125	0.531	0.435	0.435	0.268	0.125	0.194	0.268	0	0.061	0.531	0.348
21	0.348	0.531	0.531	0.268	0.636	0.268	0.531	0.348	0	0.348	0.061	0.061	0.435	0.531	0.348	0.348	0.194	0.268	0.348	0.060	0	0.435	0.268
22	0.531	0.348	0.194	0.636	0.636	0.268	0.348	0.531	0.435	0.531	0.531	0.531	0.268	0.194	0.531	0.348	0.531	0.435	0.531	0.531	0.435	0	0.125
23	0.348	0.531	0.348	0.435	0.636	0.125	0.531	0.348	0.268	0.531	0.348	0.348	0.435	0.348	0.348	0.194	0.348	0.268	0.343	0.348	0.268	0.125	0

Table 3. Dissimilarity matrix of the 23 accessions of durum wheat used in the experiment.

due to homoplasy of compound microsatellite sequences. This issue on lack of variation between some of the accessions can be further clarified by using additional microsatellite markers for the characterization. Also the UPGMA dendrogram based on SSR data (Figure 2) showed the closest correlation with the groupings of particular genotypes based on agro-morphological characters. These findings shows that genetic similarity or dissimilarity measured on the basis of molecular markers agreed with relationship measurements based on agro-morphological traits, confirming the views of Pearman (2001) and Reed and Frankham (2001). Also, these results agrees with the conclusion of Ruiz and Aguiriano (2004), that quantitative agro-morphological characters seem to be very useful to verify duplicates in a durum wheat collection because they detected differences confirmed at the molecular level and not found with the qualitative characters alone.

The results of this study indicated that SSR analysis could be successfully used for the estimation of genetic diversity among durum wheat accessions even with a small number of markers. Also these results confirm the conclusion of Plaschke et al. (1995) that a small number of markers are sufficient in detecting polymorphism among wheat genotypes. The bread wheat microsatellite markers were successfully used for estimation of genetic variation in tetraploid wheat, because of their high degree of conservation at their flanking regions. In conclusion, our data showed substantial variation in morphological traits and microsatellite DNA polymorphisms among local durum wheat germplasm. This variability among accessions is expressed in



**Figure 2.** Dendrogram showing genetic relationships among 23 durum wheat accessions constructed based on the genetic distance estimated form 3 microsatellite marker analysis. Groups A and E contain the majority of tallest and late maturing genotypes; group B contains the majority of early maturing genotypes; group C contains accessions which are mid-season maturing accession; and group D contains accessions which are short and moderately late.

differences of earliness, plant height and thousands kernel weight. The observed variability might be due to anthropogenic, geographic and environmental factors. The information on genetic diversity of local landraces is very useful for better management of Moroccan durum wheat gene pool and genetic enhancement of cultivars in durum wheat breeding programs. Further investigations are in progress for collection, conservation, characterization and utilization of Moroccan durum wheat germplasm.

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