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MORPHOLOGICAL CHARACTERIZATION OF LENTIL (*LENS CULINARIS* MEDIK.) LANDRACES FROM CASTILLA Y LEÓN, SPAIN

MARÍA D. CRISTÓBAL[1*], VALENTÍN PANDO[2], BAUDILIO HERRERO[3]

[1]*Department of Plant Production and Forest Resources, University of Valladolid, Avenida Madrid 57, 34004 Palencia, Spain*
[2]*Department of Statistic and Operation Research, University of Valladolid*
[3]*Department of Agroforestry Sciences, University of Valladolid*
[*]*Correspondence author e- mail: lcristob@pvs.uva.es*

Abstract

The characterization of plant genetic resources has a significant impact on their possible future use in lentil breeding programs and also, in the activity of germplasm collections. In this work we have characterized 27 lentil landraces from the region of Castilla and León (plateau in Northern Spain), existing among them two different morphological groups according to the characteristics of the seeds (Macrosperma and Microsperma). Fifteen morpho-agronomical characters were measured for data collection. A principal components analysis allowed the definition of the 5 factors which explain 83.7% of the cumulative variance. The first factor explains 25.3% of total variation and it is related to seed production. We performed a cluster analysis obtaining 5 groups, each one defined by the average values of the employed characters. 37.0% of landraces were included in group 5, characterized by a high mean of the harvest index (38.4) and also a high mean value (74.4) for the number of pods. To study the seeds we used a correspondence analysis. 6 dimensions which explained the 80.9% of the variance were chosen. Two cluster analysis were carried out using the coordinates of the aforementioned analysis. The aim of this work was to study the morphological variability of these populations and to discover the relationship among them.

Introduction

The lentil is one of the oldest crops used in the Iberian Peninsula. The first archaeological evidence regarding lentil consumption comes from the Argar culture, Bronze Age (Zohary & Hopt, 1994). This legume has been used by all cultures which have lived in Spain.

The lentil is a traditional legume crop in Spain, mainly cultivated in dry areas of North and South Plateau. The lentils are an important source of essential amino acids, fatty acids and trace mineral (Zia-Ul-Haq *et al.*, 2011).

In the last 50 years the cultivated area of lentil has been reduced due, in part, to low productions because it has received less attention in breeding programs than other legumes and crops. In many regions of Northern Spain the farmers cultivate lentil only for their own consumption. They sow local varieties, populations that consist of a mixture of genotypes, result of a long and lasting unconscious selection by these farmers which has theoretically resulted in a better adaptation to the land and climatic conditions of the sowing areas. These autochthonous populations have great importance since they have a lot of genes for resistance to pest, diseases and tolerance to adverse environmental conditions (Cubero *et al.*, 2006). Landraces are collected and preserved by gene banks and they represent a valuable contribution to establish the *core collection*. These collections have a representative variation of the cultivated species and in order to be able to use them in future breeding programs, they should be characterized and evaluated. Morphological characters have been frequently used in order to know the diversity in germplasm collections (Erskine & Choudhary, 1986; Sharma & Luthra, 1987; Hoffman *et al.*, 1988; Erskine *et al.*, 1989; Erskine *et al.*, 1991; Saker *et al.*, 1999; Tullu *et al.*, 2001; Poonam *et al.*, 2006; Toklu *et al.*, 2009; Tyagi & Khan, 2010; Zaccardelli *et al.*, 2012).

Barulina (1930) distinguished two groups of lentils based on differences in the weight of one hundred seeds: small seeds (less than 4.5 g) or Microsperma and large seeds or Macrosperma. Morphological variation within groups is quite remarkable. There are genetic studies that separate these two groups of lentils and associate them with morphological characters (Tyagy & Sharma, 1984; Chachota & Shrama, 1993). Mateo (1960) carried out a classification of Microsperma lentil into two varieties: *dupuyensis* known as Verdina lentil (yellow cotyledons and olive green coat with large black pattern) and *variabilis* or Pardina lentil (with orange cotyledons and brown or grey testa, with variable testa pattern and colour). Three types of lentil are cultivated in Spain (Macrosperma, Pardina and Verdina) and a quality designation for each has been created. Although they are classified in three types, any study (Bakhsh *et al.*, 2013) shows the high heterogeneity within them. Therefore, it is necessary to know the types of seed present in the different accessions. Lentil production in Spain in the last years was irregular, there have been years of high production as for example, from 2000 to 2004 (19.1 to 27.4 thousand tonnes), followed by periods of low production as from 2005 to 2010 (6.9 to 14.5 thousand tonnes) (FAOSTAT, 2012).

The aim of this work is to study the morphological characteristics, to analysis the variability and the relationships among the 27 landraces from Castilla and León (North of Spain) for their use in conservation and breeding programs.

Materials and Methods

The plant material of this study was 27 local varieties of *Lens culinaris* Medik. from Castilla y León, it is situated in the North Iberian Peninsula, and it spans a total surface of 94,147 km^2, where this legume is still cultivated. 25 samples were provided by the germplasm bank of CFR-INIA (Plant Genetic Resources Center, National Institute of Agricultural and Food Technology, Madrid) and two of them were supplied by a farmer. The weight of the samples ranged from 100 to 200 g. Table 1 shows the numbers or names given to the accessions, the town and province of origin and their belonging to one of the defined types above, as well as the original agro-climatic zone.

Characterization: The characterization was carried out in a plot in the province of Valladolid, belonging to ITACYL (Agricultural Technological Institute of Castilla and León) with coordinates 41º40'36'' N, 4º39'43'' W at an altitude of 695 m. above sea level. The trials were conducted for 3 consecutive years (2004-2006). Landraces were sown in February with a randomized block design and three replicates per variety. Each plot, 3 m long, had 4 rows at distance of 0.3 m. Sowing was done by hand every 0.1 m, 120 plants were expected per plot. Cultural operations, fertilization and weed control, were made according to local practices. The first step was ploughing with a depth among 25 to 30 cm, then, a cultivator was used. Before sowing, 300 kg/ha of fertilizer (8-15-15) (Nitrogen-Phosphate-Potassium) were added. A first weed control was done with herbicide "Gadisam" (trifluralina 24% + linuron 12%) 4 l/ha. Manual weed control and harvest was done without any additional irrigation.

Tables 2 and 3 present the list of characters used to evaluate the plants and the seeds respectively. The variables to perform the characterization were taken from the species descriptors published by AGPG (1985) and some of them were modified such as leaf area (AF), biomass (BIO) and yield (REN), or added as pod total (VT), pod length (LV), harvest index (IC) and number of leaflets per leaf (NF) with a view to improve data collection.

Table 1. Passport descriptors of lentil landraces used in this study. Region of origin: I (Mountain León), II (detritus high plateu), III (leonés high plateu), IV (limestone high plateu), average temperature of coldest month (TCM), frost period (FP) and average annual precipitation (AAP), for each region. Type of seed: (P) lentil Microsperma Pardina, (V) Microsperma Verdina and (M) Macrosperma.

Register number	Locality	Province	Region	Altitude	TCM	FP	AAP	Type
2058	Posada de Valdeón	León	I	939	7-10ºC	8-10	700-2000	P
8697	Matallana de Torío	León		1013				V
8698	Riaño	León		1048				P
8699	Riaño	León		1048				P
8688	Cistierna	León		951				V
4245	Valdepolo	León	II	805	8-12ºC	6-8	500-600	P
BAUI	Respenda de la Peña	Palencia		990				P
BAUII	Riosmenudos de la Peña	Palencia		990				P
16344	Pino del Río	Palencia		992				P
8701	Valencia de don Juan	León	III	765	8-11ºC	6-8	400-500	P
11077	La Bañeza	León		771				P
16346	Izagre	León		792				P
4246	Matadeón de los Oteros	León		858				V
11076	Sahagún	León		816				V
11083	Sahagún	León		816				P
16345	Grajal de Campos	León		800				P
4244	Grajal de Campos	León		800				P
11075	Vallecillo	León		839				P
11082	Nava de los Oteros	León		853				V
11079	Valladolid	Valladolid	IV	691	8-11ºC	6-8	400-500	M
11080	Valladolid	Valladolid		691				M
11081	Valladolid	Valladolid		691				P
11078	Valladolid	Valladolid		691				M
11074	Villanubla	Valladolid		843				M
19696	Villamuriel de Cerrato	Palencia		725				M
22153	Castrojeriz	Burgos		808				V
22154	Castrojeriz	Burgos		808				P

Table 2. Quantitative traits used in the characterization of lentil landraces.

Traits	Description
AF – Leaflets area	Average leaflets area (cm^2) in 10 plant randomly selected plants per plot
S100 – 100 seeds weight	100 seeds weight (g) three groups of 100 seeds
DF – Time to flowering	Days from snow to 50% plant have flowered
HP – Plant height	Average plant height (cm) in 10 plant randomly selected plants per plot
HV – Pod height	Average lowest pod height (cm) of in 10 plant randomly selected plants per plot
DM – Time to maturity	Days from snow to 90% plant have ripened
FPP - Flowers per peduncle	Average number of flowers per peduncle in 10 randomly chosen plant per plot
SPV – Number of seed/pod	Average number of seed per pod in 10 randomly chosen plant per plot
LV – Pod length	Average pod length (mm) in 10 pod in 10 randomly chosen plant per plot
BIO – Biological production	Biological production (g) per plot
REN – Seed yield	Total weight (g) of seed per plot
VT – Pod number	Average number of pod per plant in 5 randomly chosen plant per plot
NF – Leaflet number	Average number of leaflets per leaf in 10 randomly chosen plant per plot
IC – Harvest index	REN/BIO x 100
PN – Germination percentage	Percentage of germinated seeds sown

Table 3. Characters used to define types of seed. Adapted from AGPG (1985).

Ground colour of testa	Pattern of testa	Colour of pattern of testa	Cotyledon colour	Size seed
Brown (M)	Absent (A)	Absent (A)	Yellow (Y)	Small (G)
Green (V)	Dotted (P)	Olive (O)	Orange (N)	Large (P)
Pink (R)	Spotted (M)	Black (N)		
	Marbled (V)			

Statistical analysis: Descriptive statistics and Pearson correlation coefficient were calculated for all the bio-agronomic characters. We performed a principal component analysis (PCA) (Eriksson *et al.*, 1999) to find the relationship between the variables, in order to know which of these were associated, and which were characterizing in the same direction or in the opposite direction. The scores of the different factors obtained in PCA, stored as variables, were used for hierarchical cluster analysis, following the method of Unweight Pair Group Method with the option of Euclidean distance squared and Ward's clustering methods to maximize the difference between the groups and minimize it within them. Once the population groups were obtained, an analysis of variance was conducted, with one factor, cluster group, to detect significant differences between the groups and to determine on the basis which variables were established. Finally to compare averages of groups a test on multiple comparisons by Tukey-Kramer (Kramer, 1956) was used and allowed the classification of the groups into different categories for each variable providing difference between them. Quantitative characters were standardized and submitted to statistical analysis using SPSS programme version 15.0 (2006). With regard to the characteristics of seeds, a seed type definition was made based on the characters shown in Table 3. 100 seeds of each population were studied for these characters to observe the percentage of seed types in each landrace. The result generated a data matrix on which we carried out a correspondence analysis. With the scores obtained, we made two hierarchical analysis clusters, one for population and other for seed types, using the Euclidian distance squared and Ward's method.

Results

Morpho-agronomical characters: Table 4 presents the mean, the standard deviation and the range for metric morphological characters of the 27 accessions analyzed, providing a first approximation to the variability in the populations studied. The high variation recorded is remarkable, being more pronounced in the variables days to flowering (DF) and days to maturity (DM), probably due to climatic differences between years. The second year turned out to be the wettest year of the trial period and that it could determine the lengthening of the vegetative period. For biomass (BIO), grain yield (REN) and number of pods per plant (VT) variation is significantly higher.

Application of Pearson's correlation coefficient revealed very high correlations between VT and harvest index (IC) (0.71). VT also showed positive correlation with plant height (HP) (0.57), REN (0.54) and the number of seeds per pod (SPV) (0.50) and negatively with the weight of a hundred seeds (S100) (-0.56). S100 is negatively correlated with SPV (-0.81) and IC (-0.53) and positively with pod length (LV) (0.61).

Grain yield of the populations studied, is highly correlated to BIO (0.93), IC (0.59) and germination percentage (PN) (0.51), besides the above mentioned correlation to VT.

Table 4. Mean, standard deviation and range for morphological plant characters of lentil landraces.

Plant characteristic	Mean	Standard deviation	Range
Leaflets area (cm^2)	0.57	0.22	0.13 – 1.69
Plant height (cm)	26.02	4.96	14.0 – 43.50
Pod height (cm)	11.93	2.72	4.00 – 23.50
Time to flowering (days)	91.70	13.48	58.00 – 100.00
Time to maturity (days)	134.67	13.62	94.00 – 148.00
Flower per peduncle	2.44	0.56	1,00 – 4.00
Number of seed/pod	1.53	0.58	1.00 – 2.00
Leaflet number	12.13	1.84	7.00 – 17.00
Pod length (mm)	12.12	1.74	7.56 – 17.00
100 seeds weight (g)	3.40	1.13	2.18 – 6.87
Biological production (g)	574.85	312.89	160.00 – 1120.00
Seed yield (g)	195.01	122.78	40.24 – 476.20
Harvest index	32.78	7.52	15.99 – 44.30
Germination percentage	56.76	8.13	34.75 – 74.25
Pod number	63.67	41.34	15.00 – 193.00

Table 5. Principal component analysis: Contribution of the most important variables in each factor, % variation and % accumulated variation.

	Factors				
	F_1	F_2	F_3	F_4	F_5
Morphological character	S100 (-0.902) SPV (0.876) IC (0.730) VT (0.706)	REN (0.940) BIO (0.925)	DF (0.922) DM (0.873)	FPP (0.871) NF (0.781)	AF (0.814) HP (0.728)
% Variation	25.3	19.0	15.4	14.2	9.8
% Variation accumulated	25.3	44.4	59.8	73.9	83.7

In brackets are the factor loadings for each parameter. (S100) 100 seeds weight, (SPV) number of seed/pod, (IC) harvest index, (VT) pod number, (REN) seed yield, (BIO) biological production, (DF) time to flowering, (DM) time to maturity, (FPP) flower per peduncle, (NF) leaflet number, (AF) leaflets area and (HP) plant height

As a result of the principal component analysis, five factors were obtained, whose eigenvalues were above 1, and which explain 83.7% of the accumulated variance. Table 5 shows the most important variables for each factor and also the percentage of total and accumulated variation explained by each factor, as well as the characteristics of the crop they represent. The variables of higher weight in the first component are: number of seeds per pod (SPV), IC and VT, and they are related to seed production. S100 is the only variable that characterize in the opposite direction. This component explains 25.3% of the total variance. The second component includes 19.0% of total variation and it is characterized by BIO and REN that would be again related to the production. The third component shows that the most important variables define plant phenology (they would explain 59.8% of the cumulative variance) and are DM and DF. The fourth factor explains 14.2% and it is defined by vegetative parameters NF leaflets per leaf (NF), leaflets area (AF) and plant height (HP). The fifth factor explains 9.8% of the total variance and it is defined by parameters of flowering. Both factors would define the growth habit of the plant.

Figure 1 shows the results of cluster analysis performed with scores obtained for each landrace in the principal component analysis In this figure populations separated into 5 groups are observed. Group 1 consists of populations 11074, 11078, 11079 and 19696. These populations have in common the fact of belonging to the Macrosperma lentil type clearly separated from other groups corresponding to populations of Microsperma type seed. In group 2 there are five lentil accessions, 3 Pardina and 2 Verdina. Group 3, consists of two populations of lentil Pardina, from the same area. Group 4 shows 3 Verdina, 2 Pardina and strangely a Macrosperma population that is not clustered with the rest of them, although in data collection, S100 that is the main character to differentiate both types of lentil, did not show a weight of the seeds as high as the rest of Macrosperma, not as small as Microsperma. Group 5 is the largest, and all the lentils are Pardina except one that corresponds to a Verdina lentil.

Analysis of variance carried out to detect significant differences between clusters showed significant differences ($p<0.05$) between groups for leaflets area and germination percentage and highly significant ones ($p<0.001$) for HP, HV (pod height), LV, DF, DM, FPP (flowers per peduncle), SPV, S100, IC and VT. No significant differences were found for NF, BIO and REN. Table 6 shows the variables lead to significant differences between groups and have been subjected to a comparison of means by Tukey-Kramer test, identifying homogeneous subsets of mean that do not differ from each other. This allows us to establish different categories of each variable and describe clearly, with regard to each character, the groups obtained in the cluster. This can facilitate the breeder's work when selecting the characters and the conservation tasks view to constitute a core collection. Group 1 is characterized as low height plants and they present a late flowering and ripening. Regarding VT character, correlated with IC, it is high in groups 4 and 5, both Microsperma seed, in particular group 5 with very high IC.

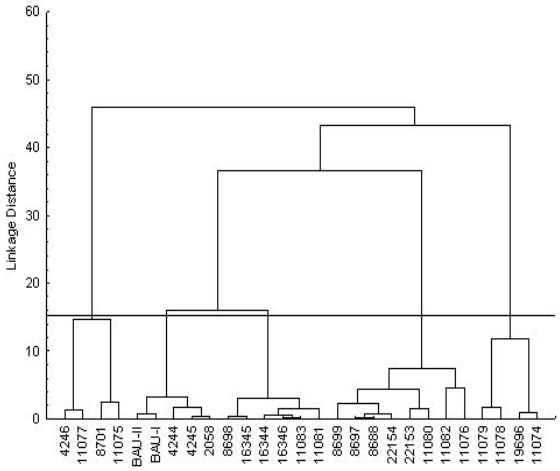

Fig. 1. Dendrogram showing five groups of populations for quantitative characters.

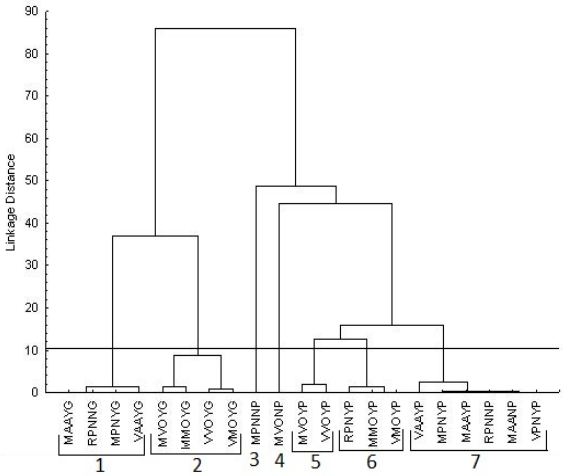

Fig. 2. Dendrogram displaying 7 groups of seeds. Group 1: Macrosperma, group 2: Macrosperma, group 3: Verdina, group 4: Verdina and Pardina (12%), group 5: Pardina and verdina (32%), group 6: Pardina and verdina (3.25%), group 7: Pardina and Verdina (5.5%).

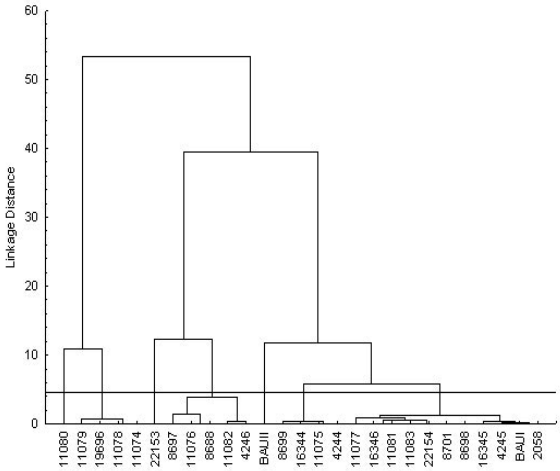

Fig. 3. Dendrogram showing seven groups of population according to the type of seed.

Seed characters: With regard to seed characteristics, 21 different types of seeds were established. They were designed by a combination of initial letters to describe the characters presented (Table 3), for example, a seed RPNNG, presents pink coat with black colour dotted pattern, orange cotyledon and large seed.

The population showing more types of seeds was 8696 with 9 different types; the lowest number was shown in population 4246 with a single type.

Table 7 shows the results of correspondence analysis applied to the matrix of 21 types of seeds and 27 populations. The number of dimensions was reduced, choosing 6 that explained 80.88% of the variance. Subsequently, the coordinates were founded in the six dimensions for rows (populations) and columns (types of seed). These coordinates were used in two cluster analysis to group the categories of variables. The dendrograms are shown in Figures 2 and 3. Groups of seeds appear in Figure 2, there are 7 groups. Group 1 consists of large seeds with orange or yellow cotyledons; the seed coat is brown, green or pink, with a dotted or plain testa pattern. Group 2, with large seeds, presents ground green or brown colour of testa and spotted or marbled pattern of testa and yellow cotyledons. Group 3, consists of a single type of small brown seed, with dotted pattern and orange cotyledons. Group 4 also consists of a single type of brown seed, with marbled pattern and orange cotyledons. In group 5 seeds are green or brown, with marbled pattern and yellow cotyledons. In group 6 ground colour of testa can be any of the identified alternatives, dotted or spotted patterns and yellow cotyledons. Group 7 has brown, pink or green seeds without pattern or with dotted pattern and cotyledons might present two colours.

Since there is great variation in the type of seed, we have assimilated insofar as possible the groups to the types of lentil defined in the introduction section, therefore, groups 1 and 2 are included in the classification of Macrosperma. Groups 4 and 5 illustrate the type of seed which forms the lentil variety Verdina, (Microsperma group) with dark background and wide pattern, which almost hides the ground of the seed, and a darker shade. Groups 3, 6 and 7 seeds have any ground of testa colour, with very slight dotted, spotted or absent pattern with a reddish-brown aspect overall appearance, and they belong to the Pardina variety (Microsperma group). Cluster analysis performed for grouping populations according to the type of seed, discriminate 7 groups (Fig. 3). Table 8 shows the frequency data of the groups of seed in each of the population groups. It is noted that population groups 1 and 2 have Macrosperma seed and 3 Verdina seed, the rest of population groups are a mixture of Pardina and Verdina in different proportions. This study of seeds reveals a great variability in their characteristics within these local varieties.

This characterization will allow, in terms of a market that demands increasingly homogeneous varieties, to make a selection of the types of lentils preferred by the consumer, in order to revalue the crop creating higher quality denominations or new ones.

Table 6. Results of the test Tukey-Kramer. Morphological characterization groups of the dendrogram. Different letters indicate significant differences, with a probability of 95%, within each column. (HP) plant height, (S100) 100 seeds weight, (HV) pod height, (DF) time to flowering, (DM) time to maturity, (FPP) flower per peduncle, (LV) pod length, (IC) harvest index, (SPV) number of seed/pod, (PN) germination percentage and (VT) pod number.

Group	HP	S100	HV	DF	DM	FPP	LV	IC	SPV	PN	VT
1	24.9^{b}	5.7^{a}	13.4^{a}	92.3^{a}	135.4^{a}	2.3^{c}	13.9^{a}	21.8^{b}	1.26^{b}	51.85^{ab}	34.9^{b}
2	24.6^{b}	2.9^{b}	11.8^{bc}	$89,9^{b}$	133.6^{a}	2.8^{a}	11.7^{b}	33.8^{a}	1.5^{a}	56.6^{ab}	62.2^{ab}
3	24.8^{b}	3.4^{b}	11.0^{b}	86.5^{c}	126.7^{b}	2.4^{bc}	10.7^{b}	28.6^{ab}	1.53^{a}	43.2^{b}	34.3^{b}
4	27.9^{a}	2.8^{b}	12.6^{ab}	91.5^{ab}	135.5^{a}	2.64^{ab}	12.4^{ab}	31.3^{ab}	1.6^{a}	59.4^{a}	76.0^{a}
5	25.7^{ab}	2.9^{b}	11.1^{b}	92.6^{a}	135^{a}	2.3^{c}	11.7^{b}	38.4^{a}	1.58^{a}	59.8^{a}	74.4^{a}

Table 7. Eigen values and inertia in the correspondence analysis with 21 variables and 27 valid cases.

Number of dimensions	Singular values	Eigen values	Percentage of inertia	Cumulative percent	Chi square
1	0.994158*	0.988351*	30.03582*	30.0358*	2668.546*
2	0.877892*	0.770695*	23.42129	53.4571*	2080.876*
3	0.512095*	0.262241*	7.96947	61.4266*	708.051*
4	0.480270*	0.230659*	7.00970	68.4363*	622.780*
5	0.464955*	0.216183*	6.56978	75.0061*	583.695*
6	0.439885*	0.193499*	5.88041	80.8865*	522.448*
7	0.402391	0.161919	4.92068	85.8072	437.180

Table 8. Percentage of the seed groups in the seven population groups. Established as a result of correspondence analysis. Brackets contain the group of seed.

Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	Group 7
68% (1) 32% (2)	94% (1) 4,74% (2) 1,25% (7)	26% (4) 74% (5)	88% (5) 9,4% (6) 2,6% (7)	22% (3) 10% (5) 68% (7)	3,25% (5) 23,5% (6) 73,25% (7)	5,55% (5) 1,55% (6) 92,91% (7)

Discussion

Obtaining the necessary date for the management of population in the genebank and determinating the degree of diversity of samples was the aim of this characterization so that they can be incorporated in breeding programs to develop cultivars resistant to adverse conditions more productive or adapted to mechanical harvesting. Considering the results of the basic statistical is observed that traits of agronomic importance, such as REN, BIO and VT have a marked variation. Toklu *et al*., (2009) also obtained large variability for characters related to production. Our data show greater diversity than the one obtained by Erskine *et al*., (1989) in a study made on a worldwide collection of lentils. Piergiovanni *et al*., (1998) found that the Mediterranean region is the one with the greatest variability in the studied traits, having analyzed populations from Africa, Asia and Europe.

According correlations, the tallest plants present a higher number of pods, a higher number of seeds per pod and seeds with a lower weigh, and these characters have contributed to high yields and high harvest index. Positive relationship between HP and REN has been proved by several authors (Hamdi *et al*., 1991; Erskine, 1996; Kusmenoglu & Muehlbauer, 1998 and Tullu *et al*., 2001). Also, as they are very tall plants they are suitable for mechanical harvesting. VT is an important character to determinate REN, as has been also shown by Bacchi *et al*., (2010) in a lentil collection from Italian and African countries, including and Microsperma types. Correlations between S100, SPV, IC and LV describe that the largest seeds are in longest pods, appearing a smaller number of seeds in these fruits. These descriptions contribute, in part, to the differentiation in two groups, Macrosperma and Microsperma (Ramgiry *et al*., 1989; Tyagi & Khan, 2010). Populations with larger seeds have a lower harvest index. The high correlation of the previous variables with BIO is also referenced by Younis *et al*., (2008) in 14 elite lines. Consequently it can be concluded that populations which produce more seeds experience more vegetative growth, with high harvest index. Results are consistent with the study made by Luthra & Sharma (1990) conducted with 56 lentil genotypes, and the one carried out by Singh (1977) with 28 cultivars of this species. Muehlbauer *et al*., (1985) considered biomass and the number of pods the major components of grain yield.

The principal component analysis shows that the first two components, which explain the highest percentage of variation are related to characters that marks the

production. Similar results were obtained by Lázaro *et al*., (2001) in a principal component analysis, in which the variables related to seed production were the most important for the first factor. The three components explain 73.1% of the cumulative variation in a Turkish lentils collection (Toklu *et al.*, 2009), value that is higher than in our study. The third component shows that the most important variables are DF and DM. The observed variation in these parameters could be used to obtain varieties of short or longer cycles, although they are very dependent of environment.

The result of hierichal cluster analysis, that will allow an improved management of the sample in the genbank, detecting those similar or even repeated, shows a separation between Macrosperma and Microsperma the latter being grouped indiscriminately Pardina and Verdina. Separation of populations based on the type of seeds is also found in lentils from Algeria, Cyprus, Egypt, Morocco, Tunisia, Pakistan and Ethiopia by Bacchi *et al*., (2010). Observing the dendrogram we can state that the varieties are not grouped considering the established climatic zones. Lázaro *et al*., (2001) did not find relationships between phenotypic and climatic characters in Spanish lentil collection, which agree with our observance. The same conclusion was achieved by Tyagi & Khan (2010) who did not find an association of 50 local varieties from India according to different eco-geographic regions, where varieties of the same area were grouped into different clusters. In our case, due to the small area of the region, it is possible that there has been an exchange of seeds among farmers in local markers. Characterization of the groups of the dendrogram shows as the Macrosperma groups is lower height than expected. This does not coincide with the results obtained in other studies (Bacchi *et al*., 2010) although in that work varieties of lentil Macrosperma improvement were included, which are much higher than Macrosperma local varieties. In our study there are only 4 not improved Macrosperma landraces. Nevertheless, they have the highest values of HV. In other studies (Karadavut, 2009) a negative correlation between S100 and HP was found which agrees with the result obtained in the present study. Lentils of the group Macrosperma have a late flowering and maturation in conjunction with the low height, leading to a lower IC as shown in Table 6, the result matches up with that obtained by Hamdi *et al*., (1991). The most productive accessions are being lentil Microsperma, most VT. Bacchi *et al*. (2010) also found that the greatest number of pods is present in Microsperma local varieties.

Conclusion

The result of this study shows the presence of a high phenotypic variation for different plant characteristics important from an agronomic point of view, allowing its use in genetic analysis and its possible inclusion in breeding programs. We have identified groups of local varieties with high yield potential. To define types of seeds and a detailed study of these in the populations have permitted to know the high diversity of seeds in these landraces, which have a mean per population of 5.11 for seed types. The data presented in this paper will also serve to identify the samples in the gene bank facilitating the work of the manager in order to identify repetitions of samples and the inclusion of others in the core collection. Furthermore, the cluster evidences that the samples are not grouped according to the origin areas and therefore they are not grouped according to determined agroclimatic characteristics, like previous studies demonstrate (Lázaro *et al*., 2001), and this will be taken into account for future strategies of sample collection.

References

AGPG. 1985. *Lentil descriptor*. International Board for Plant Genetic Resources, Rome.

Bacchi, M., M. Leone, F. Mercati, G. Preiti, F. Sunseri and M. Monti. 2010. Agronomic evaluation and genetic characterization of different accessions in lentil (*Lens culinaris* Medik.). *Ital. J. Agron.*, 4: 303-314.

Bakhsh, A., S.H.M. Iqbal and N.M. Cheema. 2013. Inheritance of morphological characters associated with plant and drief seeds in lentil (*Lens culinaris* Medik.). *Pak. J. Bot.*, 45(5): 1497-1502.

Barulina, H. 1930. Lentil of the URSS and of the other countries. *Bull. Appl. Bot. Pl. Breed Suppl*., 40: 1-319.

Chachota, R.K. and S.K. Shrama. 1993. Studies on genetic variability and component analysis in macrosperma and microsperma lentils. *Indian J. Genet.*, 53(4): 411-417.

Cubero, J.I., S. Nadal and M.T. Moreno. 2006. *Recursos Fitogenéticos*. Editorial Agrícola Española, Madrid.

Eriksson, L., E. Johansson, N. Kettaneh-Wold and S Wold. 1999. *Introduction to multi and megavariate data analysis using projection methods (PCA & PLS)*. Umetrics AB, Umea.

Erskine, W. 1996. Size effects of lentil (*Lens culinaris*) yield potential and adaptation to temperature and rainfall in West Asia. *J. Agr. Sci.*, 126: 335-341.

Erskine, W. and M.A. Choudhary. 1986. Variation between and within lentil landraces from Yemen republic. *Euphytica*, 35: 695-700.

Erskine, W., Y. Adham and L. Holly. 1989. Geographic distribution of variation in quantitative traits in a world lentil collection. *Euphytica*, 43: 97-103.

Erskine, W., A.S. Gill, A. Orhan and C. Pastrana. 1991. Heterosis in lentil. *J. Genet. & Breed*, 45: 241-244.

FAOSTAT. 2012. http://faostat.fao.org/ default.aspx?alias=faostat&lang=es (last visit, 20/02/2013)

Hamdi, A., W. Erskine and P. Gates. 1991. Relationships among economic characters in lentil. *Euphytica*, 57: 109-116.

Hoffman, D.L., F.J. Muehlbauer and G. Ladizinsky. 1988. Morphological variation in *Lens* (*Leguminosae*). *Syst. Bot.*, 13(1): 87-96.

Karadavut, U. 2009. Path analysis for yield and yield components in lentil (*Lens culinaris* Medik.). *Turk. J. Field Crops*, 14(2): 97-104.

Kramer, C.Y. 1956. Extension of multiple range test to group means with unequal numbers of replications. *Biometrics*, 12: 307-310.

Kusmenoglu, I. and F.J. Muehlbauer. 1998. Genetic variation for biomass and residue production in lentil (*Lens culinaris* Medik.). I. Relations to agronomic traits. *Crop Sci.*, 38: 991-915.

Lázaro, M., M. Ruiz, L. De la Rosa and I. Martín. 2001. Relationships between agro/morphological characters and climatic parameters in Spanish landraces of lentil. *Genet. Resour. Crop Ev.*, 48: 239-249.

Luthra, S.K. and P.C. Sharma. 1990. Correlation and path analysis in lentils. *Lens Newsletter*, 17(2): 5-8.

Mateo, J.M. 1960. *Leguminosas de grano*. Salvat, Barcelona.

Muehlbauer, F.J., J.I. Cubero and R.J. Summerfield. 1985. "Lentil" (*Lens culinaris* Medik.) In: *Grain Legume Crops*. (Eds.): R.J. Summerfield and E.H. Roberts. Collins, London, pp. 266-311.

Piergiovanni, A.R., G. Laghetti, G. Olita, M. Monti, G. Preiti and G. Di Prima. 1998. Screening for agronomic and biochemical traits in a lentil germplasm collection. In: *3rd European conference on grain legumes proceedings*. AEL (Ed.), ITACyL, Valladolid. pp. 206.

Poonam, T., K. Sharma and S.K. Mishra. 2006. Agromorphological characterization of diverse germplasm of lentil (*Lens culinaris* Medik). *New Bot.*, 23: 225-235.

Ramgiry, S.R., K.K. Paliwal and S.V. Tomar. 1989. Variability and correlations of grain yield and other quantitative character in lentil. *Lens Newsletter*, 16(1): 19-21.

Saker, A., W. Erskine, B. Sharma and M.C. Tyagy. 1999. Inheritance and linkage relationships of days to flower and morphological loci in lentil. *(Lens culinaris* Medikus subsp. *culinaris*). *J. Hered.*, 90: 270-275.

Sharma, P.C. and S.K. Luthra. 1987. Genetic divergence in lentil (*Lens culinaris* Med.). *Genet. Agr.*, 41: 349-360.

Singh, T.P. 1977. Harvest index in lentil. *Euphytica*, 26: 833-839.

SPSS. 2006. *SPSS 15.0 brief guide*. SPSS Inc., Chicago.

Toklu, F., B.T. Bicer and T. Karakoy. 2009. Agro-morphological characterization of Turkish lentil landraces. *Afr. J. Biotechnol.*, 8(17): 4121-4127.

Tullu, A., I. Kusmenoglu, K.E. Mcphee and F.J. Muehlbauer. 2001. Characterization of core collection of lentil germplasm for phenology, morphology, seed and straw yields. *Genet. Resour. Crop Ev.*, 48: 143-152.

Tyagi, S.D. and M.H. Khan. 2010. Genetic divergence in lentil. *Afr. Crop Sci. J.*, 18(2): 69-74.

Tyagy, M.C. and B. Sharma. 1984. Association of seed size with morphological markers in lentil (*Lens culinaris* Medik). *Seed Res.*, 12(2): 61-64

Younis, N., M. Hanif, S. Siddique, G. Abbas, M. Jawad Ashar and M. Ahsanul Haq. 2008. Estimates of genetic parameters and path analysis in lentil (*Lens culinaris* Medik.). *Pak. J. Agri. Sci.*, 45(3): 44-48.

Zaccardelli, M., F. Lupo, A.R. Piergiovanni, G. Laghetti, G. Sonnante, M.G. Daminati, F. Sparvoli and L. Lioi. 2012. Characterization of Italian lentil (*Lens culinaris* Medik.) germplasm by agronomic traits, biochemical and molecular markers. *Genet. Resour. Crop Ev.*, 59: 727-738.

Zia-ul-Haq, M., S. Ahmad, M.A. Shad, S. Iqbal, M. Qayum, A. Ahmad, D.L. Luthria and R. Amarowicz. 2011. Compositional studies of lentil (*Lens culinaris* Medik.) cultivars commonly grown in Pakistan. *Pak. J. Bot.*, 43(3): 1563-1567.

Zohary, D. and M. Hopf. 1994. *Domestication of plants in the Old Worl*. (2nd Ed) Oxford University Press, Oxford.

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