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Full Length Research Paper

Analysis of agromorphological diversity of southern Tunisia faba bean (*Vicia faba* L.) germplasm

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Forty two faba bean landraces (traditional farmers' varieties) (*Vicia faba* L.) from southern Tunisia, belonging to 8 oasis agro-ecosystems were assessed using morphological descriptors. The studied descriptors focused on morpho-phenological and morpho-agronomical traits. The data obtained is extracted on the basis of 35 descriptors of 42 populations from 8 localities, at a rate of 30 replications (plants) per population, in totality of 1260 individuals. The data underwent an analysis of variance and a multivariate analysis. Significant differences among populations for the thirty five descriptors were observed. The multivariate analysis performed on averages of all parameters revealed eight main groups, and variation within the same oasis agro-ecosystems was extremely important. Particularly, the group of 'Tozeur' made up of five populations (P38, P39, P40, P41 and P42), and the group of 'Medenine' composed of seven populations (P19, P20, P21, P22, P23, P24 and P25) showed a special characters than the other populations especially for seedling characters. The geographical location did not seem to be the main factor structuring the variability of the studied landraces. There proved to be substantial phenotypic variability in the Tunisian faba bean germplasm.

Key words: Diversity analysis, germplasm, faba bean (*Vicia faba* L.), oasis agro-ecosystems, Southern Tunisia.

INTRODUCTION

Broad bean or faba (*Vicia faba* L.) is the most important grain legume in the world (Lopez-Bellido et al., 2005). For decades, this legume represents the only grain legume widely grown in Europe (Gresta et al., 2009). The world production is about 26 million tons for both green and dry bean in 2008 (FAO stat, 2008). Because of its high nutritional value and its ability to grow over a wide range of climatic and soil conditions (Bond et al., 1980; Lawes et al., 1983), the faba bean is the fourth most important crop worldwide (Torres et al., 2010). By cultivation area, it ranks fourth among the cool season food legumes (2.6 million hectares per year), behind pea, chickpea and lentil(http://faostat.fao.org). Grown in the Mediterranean basin, the Nile valley, Ethiopia, Central and East Asia, Latin America, northern Europe, North America and Australia, more than 80% of the cultivation of this legume has been traditionally performed in developing countries (Torres et al., 2010).

Nowadays, its cultivation is continuously declining due to the low yield and yield instability (Gresta et al., 2009). Over the past years, many constraints have combined to limit the production and reduce the growing area of this legume. The yield potentials of existing varieties are poor, the yields are notoriously variable, predation by pests, diseases and parasites is severe, inputs are limited and production techniques are inefficient (FAO, 2008).

All these constraints caused a negative trend in maintaining the numerous old varieties present in different growing areas. This caused a strong genetic decline of the local gene pool with the real risk of losing useful genetic resources for future breeding programs and

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variety development (De Giorgeo and Polignano, 2001).

In the Mediterranean regions, this pool constitutes the basis for plant breeders to develop new varieties (Nadal et al., 2003; Terzopoulos et al., 2008). Autochthonous landraces evolved through phenotypic and empirical selection by farmers, it contains genes adaptable to different agro- ecological conditions apart from those characteristic of the Tunisian oasis ecosystems.

In Tunisia, the national average yield is lower than 50 000 tons (Ministry of Agriculture and Environment, 2009) and characterized by its instability (Khaldi and Zekri, 2002). Abiotic stresses and inefficient production techniques are the main reasons, especially in the southern parts characterized by severe aridity (Kharrat et al., 2006). Tunisian genotypes from different parts, except for those common in oasis agro-ecosystems of the country have been the subject of several studies. These studies focused on genetic (Ouji et al., 2010, 2011), biochemical (Chaieb et al., 2011), physiological (Abbes et al., 2009), phytopathological (Asma et al., 2003; Kharrat et al., 2006) and agrifood aspects. The locally adapted taxa traditionally cultivated in oases could represent an economically valuable opportunity for farmers in marginal areas of southern Tunisia where aridity, usually associated with salinity, constitutes a serious problem.

To safeguard the remaining Tunisian germplasm and to develop new improved varieties well adapted to environmental conditions of southern Tunisian areas, it was suggested that a wide germplasm collection with a large genetic diversity would be set up (Kharrat et al., 2006).

In Tunisia, the Arid Lands Institute faba bean collection constitutes a pool which is capable of conserving genetic diversity of the species. It contains a significant proportion of the Tunisian genetic resources for this species distributed in Tunisian oases. In addition, this institute contains a core collection of faba beans germplasm originating from Tunisia (Loumerem et al., 2004). In the framework of the present study, patterns of morphological diversity were examined in relation to the geographical origins of 42 populations of *V. faba* L. from 7 agro-ecosystem oases. This study was conducted to investigate whether the faba bean core collection was well defined. It aimed at estimating the pattern of genetic diversity among the Tunisian arid region faba bean landraces, the relative contribution of various morpho-agronomic traits to the total diversity in the germplasm and to study the relationship between the geographical origin and genetic diversity.

MATERIALS AND METHODS

Plant material origin

During 2006, a total of 42 populations of local faba bean seeds (*Vicia faba* L.) were collected and maintained at the experimental field of the Arid Lands Institute of Medenine (Tunisia). The code number, local cultivar´s name, oasis name and province of each entry are recorded in Table 1.

The prospected areas cover the totality of agro-ecosystem oases in the Southern Tunisian regions of Gabes, Gafsa, Medenine, Tataouine and Tozeur. Seeds were collected from each region from different farmers. At the same time, a semi structured interviews were carried out with farmers who donated the samples at each collection site to take down detailed information on local cropping practices.

Experiments and experimental design

The present study was conducted in the Arid Lands Institute (Medenine, Tunisia) located in the lower arid bioclimate (Table 2). Collection including this species was initiated and plants were propagated in the experimental field.

The experimental design consists of a randomized complete block laid out on the experimental field, and field trials consisted of 120 m^2 measurement for one plot. Each entry consisted of three plots with ten plants per plot. Each plot consisted of 42 rows (number of populations) 10 m long with a spacing of 40 cm between rows and 20 cm between plants. Thus, the total number of plants studied per population was thirty. Sowing began on 15^{th} November 2007. No fertilizers or any other soil additives were applied. Plants were surface-irrigated at regular 2 weeks intervals periodically.

Measurements

Thirty plants randomly selected from each entry (populations) were considered to measure the 35 quantitative traits indicated in Table 3. The studied traits were focused on morpho-phenological aspects and morpho-agronomic characteristics [according to the ``Bioversity international´´ and UPOV (2003) descriptors]. Some other characteristics were considered according to UPOV approval for examination of Distinct, Uniform and Stable (DUS).

Statistical analysis

Simple descriptive statistics (such as means and coefficient of variation) were used in order to compare variation between the studied populations. A variance analysis (ANOVA) was performed and then the averages were compared by least significant difference (LSD) test. On each parameter, a correlation analysis was then used to estimate the relationship between the studied variables. The 42 populations were clustered based on phenotypic traits. The scales portray a dissimilarity index calculated using the Euclidean distance coefficient and the dendrogram was developed using unweighted pair group method using arithmetic averages (UPGMA) clustering procedures. In order to classify the populations and to identify the most important traits, multivariate analysis methods was used: the principal component analysis (PCA) and the multiple-component analysis (PCM). All calculations were made using SPSS (16.0) and Xlstat (2009) statistical software.

RESULTS

Population variability and agro-ecosystem effect

The range of the measurements, the average, and the coefficient of variation for the continuous characteristics are shown in Table 4. The data show large variability

Table 1. Locations and characteristics of seeds of *V. faba* L. populations collected from the oases of southern Tunisia.

Pop	code	Oasis agro-ecosystem	Oasis name	Province	Latitude (N)	Longitude (E)	Altitude (m)
1 to 8	P1	Oasis of Mareth (8 populations)	Mareth	Gabes	33°37'	10°16'	48
	P8				33°37'	10°16'	48
9 to 25	P9	Oasis of Médenine (17 populations)	Medenine	Medenine	33°20'	10°29'	48
	P25				33°20'	10°29'	48
26 to 32	P26	Oasis of Beni Khedache (7 populations)	Beni Khedache	Medenine	33°15'	10°11'	506
	P32				33°15'	10°11'	506
33	P33	Oasis of Gafsa (2 populations)	Tafartassa	Gafsa	34°24'	8°46'	294
34	P34				34°24'	8°46'	294
35	P35	Oasis of Tataouine (1 population)	Ferch	Tataouine	32°55'	10°27'	235
36	P36	Oasis of El Hamma (2 populations)	El Hamma	Gabes	33°52'	9°47'	64
37	P37				33°52'	9°47'	64
38 to 42	P38	Oasis of El Hamma (5 populations)	Tozeur	Tozeur	33°55'	8°08'	43
	P42				33°55'	8°08'	43

Table 2. Trial site characteristics in El Fje (I.R.A Medenine).

Location	Site	Mean temp. min °C (winter)	Mean temp. max °C (summer)	Mean rainfall	Soil	Water quality
Medenine	El Fje	10	30	150 (mm)	Sandy	Tap water

between landraces for all traits. The differences of the mean values for all the continuous morphological characteristics were statistically significant at 5% (Table 4). Number of stems per plant showed the highest coefficient of variation (58.25%) followed by 1000 seeds weight descriptor (43.05%), number of flower per raceme (42.14%), pod length (37.85%), pods per truss (33.76%) and ovules per pod (37.62%). The lowest coefficient of variation was observed for time of maturation of the first pod (04.16%) and flower length (09.80%). Coefficients of variation showed a lot of variation between populations for some characters, especially in those related to yield.

The important mean value of stems per plant reported in populations from Beni Kedache agro- ecosystem (7 populations) established an average of 4 stems. The populations of Medenine agro-ecosystem had the highest number of flowers per raceme (4 flowers) and number of pods per truss (1.19 pods). Tozeur agro-ecosystem populations have the longest pods with a mean value of 16.71 cm and the highest number of ovules per pod with 5.40 ovules. The highest average of 1000 seeds weight was observed for populations from Tataouine agro-ecosystem with 1827 g. All other traits relating to leaflet and flower shape evidenced less variable mean values.

Least significance difference (LSD) test ($P<0.05$) showed the existence of two variable categories. The first one seems to be more discriminative; this class of descriptors comprised eight quantitative variables: Days to flowering, plant height, stems per plant, flowers per raceme, the pod length and width, and number of ovules per plant (including seeds). The 1000 seeds weight remains the most discriminate descriptor; it can subdivide the germplasm only into three groups (minor, equina and major Var). When we consider this category of descriptors, populations from Tozeur agro-ecosystem (populations 38, 39, 40, 41 and 42) are clearly separated from the other populations' values in size of pods, maximum length and width, and number of ovules per plant. The populations 19, 20, 21, 22, 23, 24 and 25 from Medenine agro-ecosystem have the lowest size or shape of pods and weight of seeds, but the highest for the number of flowers per raceme. Also, it is noticeable that the population of Mareth (P1 and P8) presents the

Table 3. Agronomical characteristics used for the description of the faba bean populations.

Continuous (agronomic) characteristics	**Code**	**Scale**
Days to flowering	DFF	(days)
Days 50% flowering	DHF	(days)
Plant height	PH	(cm)
Stem thickness	ST	(mm)
Stems per plant (including tillers more than half the length of the main stem)	SPP	------
Nodes per stem	NPS	------
Leaflet length (basal pair of leaflet at secondary node)	LL	(cm)
Leaflet width (basal pair of leaflet at secondary node)	LW	(cm)
Flowers per raceme	FPR	------
Flower length	FL	(cm)
Pods per truss	PPT	------
Pod length	PL	(cm)
Pod width (from suture to suture)	PW	(mm)
Ovules (including seeds) per pod	OPP	------
1000 seed weight	SW	(g)
Time of full development of pod (first fully developed pods)	TFDP	(days)
Ordinate (morphological) characteristics	Code	Scale
Homogeneity of plants	HP	3, 5, 7
Growth type of plants	GTP	1, 2
Anthocyanin coloration of stem	ACS1	1, 9
Greyish hue of green color of leaves	GHGL	1, 9
Intensity of green coulour on leaves	IGCL	3, 5, 7
Position of maximum width leaflet (basal pair of leaflet at secondary node)	PMWL	1, 2, 3
Leaflet folding (along the main vein, terminal pair of leaflets)	LF	3, 5, 7
Melanin spot of wing	MSW	1, 9
Colour of melanin spot of Wing	CMSW	1, 2, 3
Melanin spot of standard	MSS	1, 9
Anthocyanin coloration of standard	ACS2	1, 9
Extent of anthocyanin coloration of standard	EACS	3, 5, 7
Pod attitude	PA	1, 3, 5, 7, 9
Degree of curvature at green shell stage of pod	DCGP	1, 3, 5, 7
Intensity of green color of pod	IGCP	3, 5, 7
Thickness of pod wall of pod	THWP	3, 5, 7
Shape of median longitudinal section of dry seed	SSDS	1, 2, 3
Color of testa (immediately after harvest) of dry seed	CTDS	1, 2, 3, 4, 5
Black pigmentation of hilum of dry seed	BHDS	1, 9

Table 4. Mean square, CV% and significance (LSD test) of morpho-agronomic characters of 42 faba bean populations from southern Tunisia.

Trait	Oasis							Combined			
	Mareth	Medenine	Beni Khedache	Gafsa	Tataouine	El Hamma Gabes	El Hamma Tozeur	Mean	C.V (%)	LSD	Sig.
Days to flowering	62.17 ± 4.03	66.35 ± 9.51	63.48 ± 2.56	67.00 ± 3.08	61.67 ± 0.48	64.33 ± 1.71	64.68 ± 3.93	64.69 ± 6.78	10.48	14.71	< 0.0001
Days of (50%) flowring	69.47 ± 4.15	74.09 ± 10.65	70.48 ± 3.85	72.00 ± 1.84	63.67 ± 1.27	68.33 ± 2.65	73.23 ± 2.47	71.87 ± 7.64	10.62	23.01	**
Stem thickness	07.34 ± 2.02	07.14 ± 2.07	06.73 ± 1.41	06.09 ± 1.58	06.50 ± 1.69	06.80 ± 1.46	07.94 ± 1.83	07.12 ± 1.92	26.93	10.63	**
Plant height	55.38 ± 12.11	51.47 ± 11.61	51.66 ± 10.42	46.27 ± 10.16	46.13 ± 8.59	45.38 ± 12.70	52.28 ± 11.05	51.67 ± 11.64	22.52	10.80	**
Stems per plant	03.66 ± 1.71	03.41 ± 2.05	04.00 ± 2.17	02.82 ± 1.36	02.77 ± 1.48	02.62 ± 1.70	02.60 ± 1.75	03.38 ± 1.97	58.25	11.55	**
Nodes per stem	21.63 ± 4.39	21.11 ± 4.74	21.62 ± 3.88	18.83 ± 4.26	19.33 ± 3.92	19.95 ± 4.21	20.26 ± 4.08	20.98 ± 4.45	21.20	5.98	**
Leaflet length	06.33 ± 1.41	06.15 ± 1.32	06.34 ± 1.30	05.70 ± 1.00	06.07 ± 1.30	06.14 ± 1.43	06.74 ± 1.41	06.27 ± 1.35	21.60	5.97	**
Leaflet width	02.93 ± 0.76	02.92 ± 0.72	02.75 ± 0.73	02.48 ± 0.61	02.77 ± 0.60	02.42 ± 0.72	03.11 ± 0.76	02.87 ± 0.74	25.91	11.34	**
Flowers per raceme	03.06 ± 0.99	04.00 ± 1.75	03.09 ± 1.11	02.80 ± 1.12	01.93 ± 0.87	03.42 ± 1.00	03.52 ± 1.12	03.47 ± 1.46	42.14	27.07	**
Flower length	02.87 ± 0.30	02.81 ± 0.24	02.90 ± 0.29	02.50 ± 0.00	02.83 ± 0.24	03.14 ± 0.38	02.97 ± 0.14	02.86 ± 0.28	09.80	40.74	**
Pod length	08.64 ± 1.91	08.00 ± 2.27	08.51 ± 2.02	09.08 ± 1.67	11.23 ± 2.24	10.07 ± 2.95	16.71 ± 3.25	09.47 ± 3.59	37.85	290.39	**
Pod width	19.32 ± 2.61	17.47 ± 3.82	19.45 ± 2.66	18.50 ± 2.39	19.97 ± 2.19	18.50 ± 2.75	20.28 ± 2.96	18.65 ± 3.35	17.96	22.87	**
Pods per truss	01.17 ± 0.40	01.19 ± 0.44	01.13 ± 0.35	01.15 ± 0.36	01.10 ± 0.31	01.07 ± 0.25	01.12 ± 0.33	01.16 ± 0.39	33.76	1.81	**
Ovules per pod	02.57 ± 0.65	02.96 ± 0.79	02.71 ± 0.74	03.13 ± 0.72	03.07 ± 0.58	03.60 ± 1.30	05.40 ± 1.18	03.18 ± 1.20	37.62	213.79	**
1000 seed weight	1735.7 ± 56.69	1354;0 ± 77.08	1596.2 ± 50.68	1426.9 ± 55.29	1827 ± 37.08	1439.7 ± 44.92	1564.2 ± 50.33	1511.5 ± 65.10	43.05	12.47	**
Time of full development of pod	164.82 ± 5.93	167.60 ± 8.66	169.71 ± 5.26	172.17 ± 3.42	169 ± 2.99	167.83 ± 3.42	170.56 ± 4.19	168.04 ± 7.00	04.16	19.11	**

Means with different letters in the same trial line differ significantly according to LSD's test (*P*<0.05); (< 0.0001) mean that Pr < F: significant (**). CV, Coefficient of variance; Sig., significance.

highest length of plant and 1000 seeds weight.

Discrimination method

Based on the combination of the results from the Neighbor-joining method (Figure 1) and PCA analysis for all the characteristics (Figure 2), the populations constituting one group could be divided into three sub-groups (Table 5). The application of PCA on the category of continuous parameters showed that the minor faba bean populations of Medenine were projected to the left of axis 1; with two groups (IA and IC) together (Figure 2, areas A and C). For group II, on the basis of PC-2 axis, the populations fall together (except population 38) in area C and D (Figure 2). Finally, the majority of populations of groups III (IIIB and IIID) can be found separated into areas B and C, except for populations 10 and 39 forming group IIIA which placed in area A. Groups I (A and C), II (A, C and D) and III (A, B and D) are seen in Table 5 with the exception of population 38 which could therefore be separated on the basis of PC-1 and PC-2 (Figure 2).

The coefficient of the various traits with values greater than 10 (Table 6) provides an indication as to which traits play a major role in the placement of the populations in the eight groups (Table 5). Along the PCA-1 axis, the related traits are stem thickness, plant height pod width and leaflet width, leaflet length, nodes per stem, while for PCA-2 axis, the corresponding traits are days to flowering, days to 50% flowering, pods per truss, 1000 seeds weight and leaflet length and width. This grouping is essentially to justify and better identify the results of Table 6. The third axis or PC-3 is defined by three additional characters; stems per plant, number of ovules (including seeds) per pod and time of full development of pod.

To verify the discrimination role of qualitative characters in the classification of populations of beans, we proceeded to the multiple-component analysis of 19 qualitative characters for 42

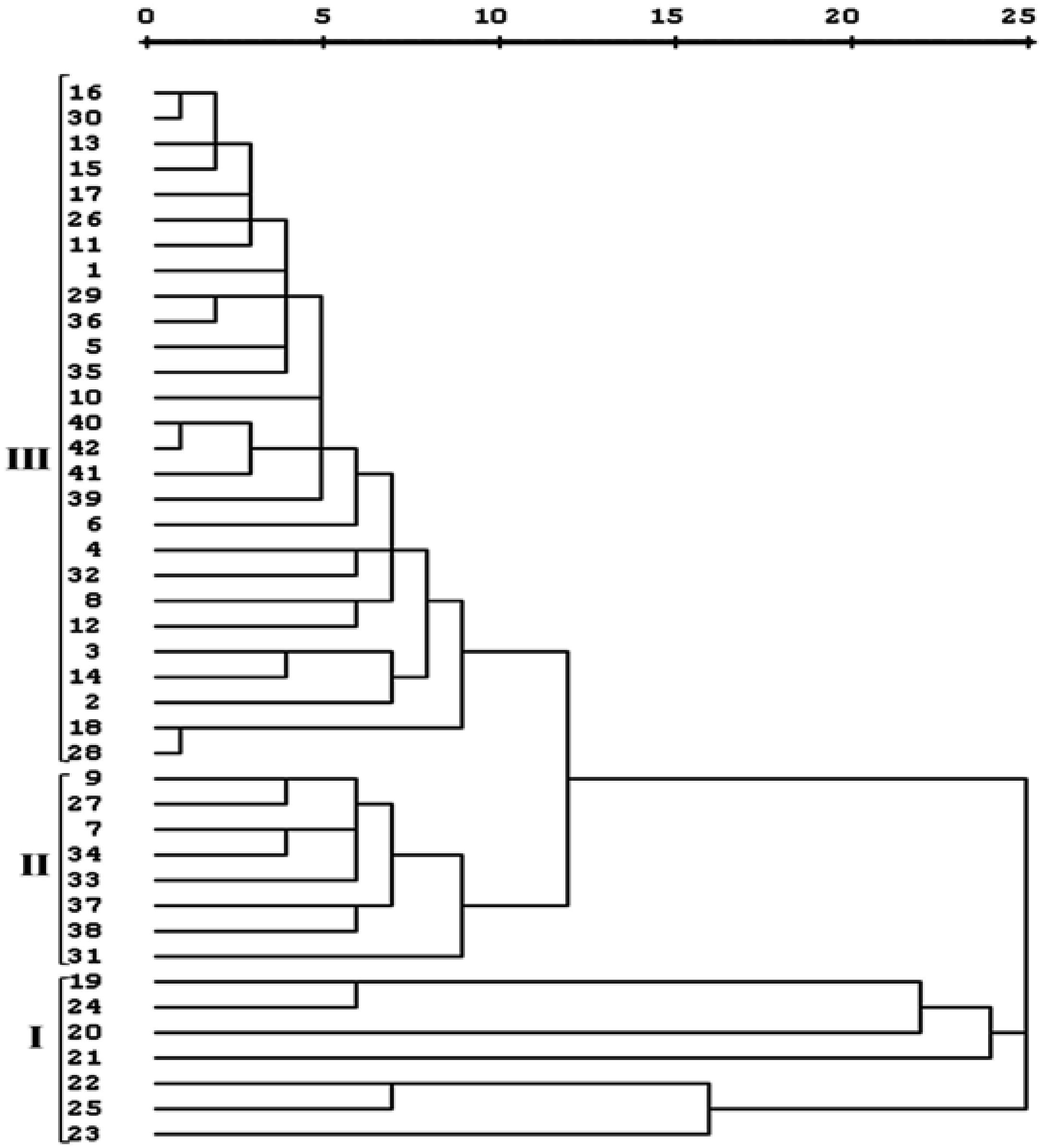

Figure 1. Dendrogram of 42 populations of faba bean from southern Tunisia based on sixteen characteristics using Neighbor-joining method.

populations of beans. Using this analysis (PCA), results were confirmed by the isolation of populations of Tozeur (P38, P39, P40, P41 and P42) and populations of Medenine (P19, P20, P21, P22, P23, P24 and P25) with current weight of other populations (Figure 3). It was revealed that the genetic diversity of populations of faba bean grown in the arid Tunisian region was not evenly distributed in all areas and some geographical structure is possible.

DISCUSSION

In the south of Tunisia, the arid climate, the failure and mismanagement of irrigation systems, especially with poor water quality of irrigation and inappropriate farming practices have led to cause the erosion of many cultures, such that the salinity in these regions has reached critical levels limiting the yield of most crops, and especially for faba bean crops. The evaluation of *ex situ* germplasm collections of crop landraces (traditional farmers' varieties) is crucial to the utilization of genetic resources for crop improvement (Maxted et al., 2003). So, it is necessary to seek the passport data describing the sites where landraces are collected, and along with this, the genotypes identity. This provides basic data on accessions in germplasm collections (Redden et al., 2007). In the principal Mediterranean productive areas

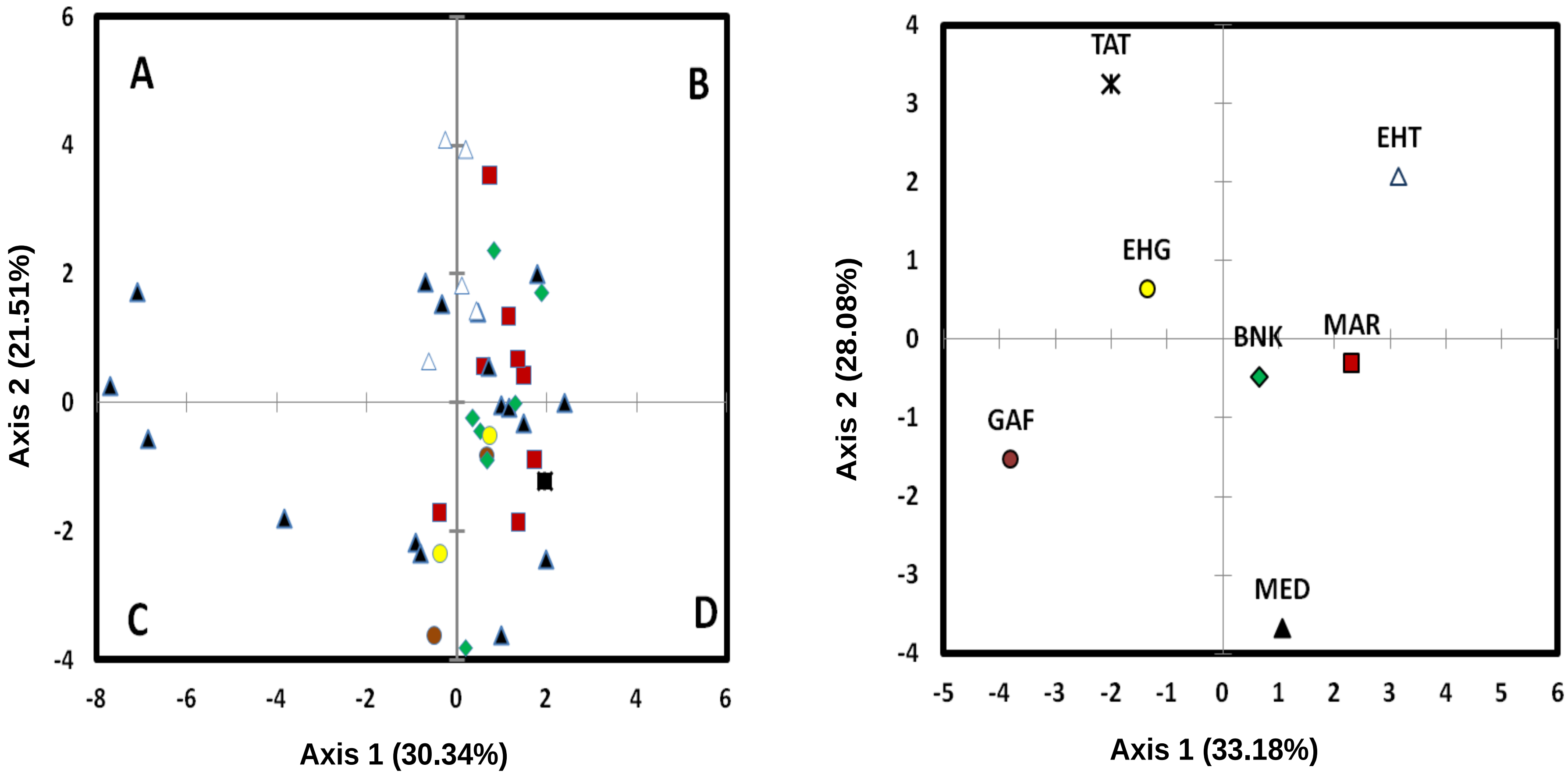

Figure 2. The Principal components analysis of forty two populations based on the sixteen characteristics. Codes indicate populations and oasis of origin: (■) Mareth, (●) El Hamma (Gabes) , (●) Gafsa, (△) El Hamma (Tozeur), (▲) Medenine, (◆) Beni Khedache and (Ӿ) Tataouine.

Table 5. The final assignment of the forty-two faba bean populations to the eight groups on the basis of the combination of the Neighbor-joining and PCA methods, based on the sixteen morphological traits.

Group name	Identified group							
	IA	IC	IIA	IIC	IID	IIIA	IIIB	IIID
Population code	20, 22, 25	19, 21, 23, 24	38	7, 31, 37	9, 27, 33, 34	10, 39	1, 4, 5, 6, 8, 11, 12, 17, 18, 26, 28, 35, 40, 41, 42	2, 3, 13, 14, 15, 16, 29, 30, 32, 36

Table 6. The principal component coefficients (PC-1, PC-2 and PC-3) based on the sixteen continuous traits.

Principal component (axes)	PC-1	PC-2	PC-3
Eigen value	5.309	4.492	3.623
% variance	33.182	28.076	22.644
Cumulative	33.182	61.258	83.902
Character	**Eigen vector**		
DFF	-0.976	-10.416	13.017
DHF	3.012	-10.729	8.179
ST	16.610	0.307	1.891
PH	14.958	-1.341	-1.814
SPP	2.393	-4.831	-12.550
NPS	11.027	-2.758	-5.745
LL	15.534	2.601	0.722
LW	13.661	0.220	0.033
FPR	5.421	-8.762	5.343
OPP	2.206	4.297	18.994
PPT	1.905	-10.816	-0.865
FL	4.427	3.351	0.004
PL	2.344	10.084	10.891
PW	1.414	16.822	-0.139
SW	0.151	12.376	-9.090
TFDP	-3.960	0.287	10.723

DFF, Days to flowering; DHF, days 50% flowering; ST, stem thickness; PH, plant height; SPP, stem per plant; NPS, nodes per stem; LL, leaflet length; LW, leaflet width; FPR , flowers per raceme; OPP, ovules (including seeds) per pod; PPT, pods per truss; FL, flower length; PL, pod length; PW, Pod width; SW, seed weight; TFDP, time of full development of pod.

(Spain, Turkey, Syria, Italy, Morocco…etc), several morphological studies have been published to assess the variation of several traits on the evaluation and characterization of broad bean populations (Perrino et al., 1991; Polignano et al., 1999; Terzopoulos et al., 2003; Terzopoulos et al., 2004).

In the present study, focusing on Tunisian faba bean germplasm, there were highly significant differences among the populations for most of the traits at several locations indicating the presence of adequate variability among the populations. Based on LSD's test, we found that the source of variability of these populations was related to sixteen quantitative characters; moreover, only eight characters give a good discrimination of the studied populations. The parameters which showed the highest coefficient of variation and with powerful discriminative effect are: 1000 seeds weight, number of flowers per raceme, number of pods per truss, pod width, number of ovules per pod (including seeds) and stems per plant. This result was considered as the first step towards choosing the most suitable discrimination characters of our populations and to illustrate the degree of variability of studied characters.

According to Terzopoulos et al. (2003), the quantitative and agronomic traits are very important in the evaluation and characterization of populations of broad bean. Terzopoulos et al. (2003) affirm that the comparison of the agronomic traits relating to yield of Tunisian populations with others cultivars shows many differences. Concerning the plant height trait, for example, populations from North Africa are the highest among a total of 1565 entries of faba bean maintained in the Bari world collection (Polignano et al., 1999). According to Terzopoulos et al. (2003), Tunisian populations are higher than the Greek ones (51.67 and 39.04 cm respectively) and smaller than Ethiopian and Turkish populations (Keneni et al., 2005; Alan and Geren, 2007). However, the coefficient of variation for the Tunisian germplasm extracted, on the basis of agronomic traits, is more important (0.225) than the coefficient found in the Ethiopian genotypes (0.086) (Keneni et al., 2005).

In the framework of the present study, it has been demonstrated that the number of flowers per raceme of the southern Tunisian broad bean ranged from 1 to 13 with an average value of 4; a very significant number in comparison with those found by Terzopoulos et al. (2003) (from 1 to 7; average 2.68), Terzopoulos et al. (2004) in Greece (from 2.5 to 3) and by Polignano et al. (1999) in North America and North Europe where four or five flowers are frequent. This difference is also observed for the coefficient of variance, which is 0.442 in southern Tunisian material and 0.329 in Greece genotypes

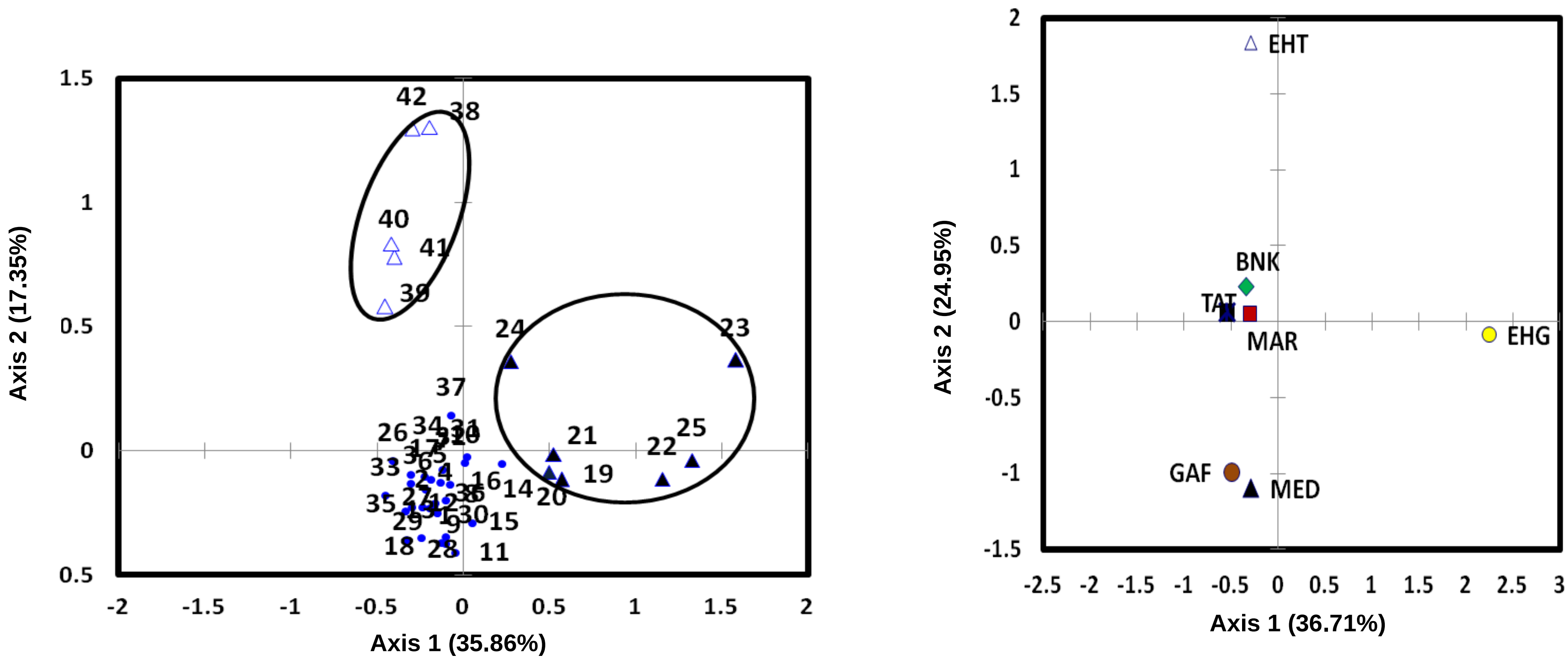

Figure 3. The Multiple Components Analysis of forty two populations based on the nineteen qualitative characteristics.

(Terzopoulos et al., 2003).

Generally, faba bean crops in Mediterranean conditions are always sown in autumn and often on rained lands. Rainfall is the most influential environmental factor (variations in rainfall give rise to variations in yield, curtailing yield at high plant densities due to increased water requirements) (Stringi et al., 1986), and it is known that the faba bean is susceptible to drought between the flowering (Duc, 1997; Aguilera-Dıaz and Recalme-Manrique, 1995) and fruiting period (Ruggiero et al., 1999). In the present study, the period from sowing to flowering varied from 55.33 to 87.66 days, and to maturity from 151 to 178.66 days in the arid condition of Medenine. The period of flowering is more important (55.3 to 87.66) than those shown in previous studies [30 days for Anouar-Sadli et al. (2008), from 22 to 33 days for Benachour et al. (2007) and 53.28 days for Keneni et al. (2005)].

The period of maturity for *V. faba* populations (151 to 178.66 days) cultivated in the south of Tunisia, is situated in middle position once it has been compared with values found in previous studies (Amanuel et al., 2000; Keneni et al., 2005; Anouar-Sadli et al., 2008). Populations of southern Tunisia are higher in stem thickness (7.12) than those found by Terzopoulos et al. (2003) in Greek germplasm (2.17).

On the whole, the single pod carries 2 to 10 ovules (Duc et al., 1999; Link et al., 2005); in our study, the populations are similar with regard to pod length (9.47 cm) and number of ovules per pod (3.18) to those presented by Terzopoulos et al. (2003) in Greece (9.19 cm and 3.52). On the basis of a core collection taken from the world faba bean collection maintained at C.I.F.A., Córdoba, Spain and under different selling conditions of yield traits in *V. faba* major, equina and minor botanical groups for two years, Nadal et al. (2003) found a pod length mean ranging from 5.33 cm (minor Var.) to 13.04 cm (equina

Var.).

According to Stringi et al. (1986), in Mediterranean conditions, the faba bean crops are always sown in autumn. Duc (1997) confirmed that winter cultivars usually have higher branching (4 to 6 stems/plant) than spring ones (1 to 2 stems/plant). For this reason our populations may be classified in the “winter” class, having a mean ranging from 1.32 to 6.43 stems per plant.

The average of number of pods per cyme (1.16) seems very small compared to the result reported by Keneni et al. (2005) in Ethiopia (6.42). The character weight of 1000 seeds seems to be the most important trait. A number of authors are interested in this character for several reasons. It is considered as the main character in the classification of *V. faba* L. We have identified several results presented by Keneni et al. (2005) in Ethiopia (423.14 g), Alan and Geren (2007) in Turkey for both cultivars (1155.2 g (Tarzan cultivar) and 1386.8 g (Eresen-87 cultivar)) and Abbes et al. (2007) in Tunisia also for both cultivars in both regions [baraca cultivar (606.5 g in Beja and 688.8 g in Oued Meliz), Bader breeding line (459.3 g in Beja and 539.8 g in Oued Meliz)].

This character ranges from 441.19 to 2140.73 g (based on populations classification) and from 1354 to 1827 g (based on oases classification) with an average of 1511.52 g for populations from the south of Tunisia. This result is in agreement with those quoted by Duc et al. (2010) that have limited the size of 200 to 2600 g per 1000 seeds. The weight of seed populations of the Southern Tunisia remains the largest on average compared to other international varieties.

Duc (1997) mentioned that the widespread cultivation of *V. faba* L. with a 1000-seeds weight of 1 kg (*V. faba* L. major) is localized in the countries of the Southern Mediterranean and China, while medium seeded types (*V. faba* L. equina) have developed throughout the Middle East and North Africa particularly in Egypt.

The results of the application of UPGMA, Neighbor-joining and PCA on all sixteen characteristics showed three groups of populations (group I, from Medenine, this group could be close to minor Var: populations 19, 20, 21, 22, 23, 24 and 25). The second group (probably belonging to equina Var.) comprised nine populations from different regions, 3 populations from beni khedache (P27, P33 et P34), 2 populations from Gabes, P7 (from Mareth) and P37 (from El Hamma), one population from Médenine (P9), one from Gafsa (P31) and the last from Tozeur (P38). The populations of this group are characterized by mean values for almost all the characters and especially of the weight of 1000 seeds. The last group, which could be related to major Var, is composed of a mixture of populations (27 populations) from different regions. It is clear that populations from Tozeur were well isolated from the remaining populations, and they had the most important characters of this group (Figure 2). These populations are distinguished from others by a very large pod size (length and width of pod) and by the highest number of seeds per pod (from 6 to 8 seeds per pod).

The multiple components analysis, based on the nineteen qualitative characteristics of forty-two populations, had distinguished, also, the minor Var. and Tozeur groups from others. Therefore, the qualitative characters confirm the results of quantitative characters.

The values of morphological traits obtained from populations from five regions in Southern Tunisia show two distinguishable groups: The group of Tozeur made up of five populations (P38, P39, P40, P41 and P42), and the group of Medenine composed of seven populations (P19, P20, P21, P22, P23, P24 and P25) forming the class of minor populations.

The data obtained in this study showed the important variability of the faba bean as regards to shape pods and seeds collected from populations cultivated in seven different oases in the south of Tunisia. These results suggest that the importance of preserving the genetic resources of the faba bean and could be a starting point for further studies. These new ‘‘populations’’ would be viewed as a kind a ‘‘core collection’’ which is maintained in the gene bank and in the field, with the aim of recurrent selection with highest yield and quality for intensive cultivation or with particular qualitative characteristics for improved food and feed uses. Moreover, the development of varieties more adapted to actual conditions (drought and salt stress) must take into consideration a strategy that prevents genetic erosion as the reduction of the biodiversity of this species in this region is a real risk. Thus, evaluation of the biodiversity of the faba bean dispersed throughout the region of Southern Tunisia is a fundamental step for the implementation of a conservation strategy. However, it is of course necessary to maintain the initial populations as a reserve collection so that all initial variation is preserved. For the faba bean, Toker (2004) reported that its heritability was affected by the type of genetic material and yield level of environment due to the fact that the plant height, number of stems and pods per plant, seed yield, biological yield, 1000 seed weight, days of flowering and maturity of plants were influenced by the effects of the both genes and environment. Further, many other environmental factors influence the improvement of faba bean culture. Plant density and early sowing were considered to be among the most important factors in the cultivation of beans. This factor can influence the number of nodes per plant (Barry and Storey, 1979) and pods per plant (Thompson and Taylor, 1977), and consequently, on the performance or yield of the faba bean (Thompson and Taylor, 1977; Kambal, 1969; Yassin, 1973). Bellido-Lopez et al. (2005) found that the number of stems per plant appears to be the most influential factor, although further research is needed to confirm this.

Although, the morphological characterization of the different surveyed populations of southern Tunisia did not help to distinguish diversity among any other populations, especially the Mediterranean populations, some structuring between and within geographical populations seem to exist. Also, the use of other biochemical and molecular markers such as isozymes (Ouazzani et al., 1995), seeds proteins (Liao et al., 1997) and random amplification of polymorphic DNA (RAPD) fingerprints (khadari et al., 1995; Trifi et al., 1998) could supply complementary useful information about faba bean germplasm in Tunisia.

In this research, using morphological data, the genetic diversity of faba bean germplasm in southern Tunisian area was revealed. The 42 studied accessions could be placed into three groups. The univariate and multivariate analysis, applied on data relating to the Tunisian faba bean material and based on 35 descriptors did not reveal any ecogeographic effect on the population average, or any population geographical area interaction. That meant that observations for all ecogeographical zones could be grouped to assess the population effect. In addition, it is interesting to note that Tunisian broad bean material shares a fairly large amount of similarity with respect to three different varieties: Equina, minor and major.

Tunisian accessions, in fact, were clustered together with landraces probably belonging to different varieties common in the Mediterranean area (*equina*, *minor* and *major* Var.). However, a substantial level of genetic variability still exists with Tunisian accessions as detected by phenotypic analysis with the possibility of introgression between varieties. This vast genetic diversity can be used in breeding programs in order to investigate the advantageous adaptation potentialities of this material.

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