

Study of Genetic Variability of Natural Population of *Allium roseum* L. (*Alliaceae*) In Tunisia

^{1,2}Guetat Arbi, ²Boussaid Mohamed and ¹Neffati Mohamed

¹Institut des Régions Arides 4119 Médenine, Tunisie

²Institut National des Sciences Appliquées et de Technologie, B.P. 676, 1080 Tunis Cedex, Tunisie

Abstract: Twenty eight populations of *Allium roseum* plants (*Alliaceae*) in Tunisia, belonging to 5 bioclimatic stages, were characterized with seven reproductive vigor descriptors: Number of Seeds produced per plants (NS), Number of Small Bulbs (NSB), Number of Mean Bulbs (NMB), Number of Big Bulbs (NGB), Total Number of Bulbs (TNB), Seeds Weight (SW) and Weight of Total Bulbs (WTB). The data underwent an analysis of variance and a factorial discriminant analysis and the Mahalanobis distances between the populations were calculated. Significant differences existed between the populations for the 7 descriptors and the discriminant analysis revealed 4 main groups among the Tunisian *Allium roseum* plants. The populations in the 5 bioclimatic stages were not homogeneous and bioclimatic stages of origin did not seem to be the main factor structuring the variability of the study populations. There proved to be substantial genotypic variability in the Tunisian material for the vigor descriptors. Variation within the same bioclimatic stage was extremely important.

Key words: *Allium roseum*, genetic variability, reproductive vigor, Tunisia

INTRODUCTION

The genus *Allium* L. (*Alliaceae*) exhibits a great diversity in various morphological characters, particularly in life form (bulb or rhizome) and ecological habitat. It is of major economic importance as vegetable crops, herbal crops and ornamental plants (Ricroch *et al.*, 2005). This genus consists mostly of perennial and bulbous plants and it is widely distributed over Holarctic regions from the dry subtropics to the boreal zone (Stearn, 1992).

Allium roseum L. (*Alliaceae*) is a species with a typically Mediterranean distribution. It belongs to section *Molium* and comprises different intraspecific taxa. This species was used in traditional pharmacopoeia for their expectorant properties and it so heavily threatened (Marcucci and Tornadore, 1997). Nevertheless, during its collects even bulbs were arrached. This spontaneous geophytic species was extensively widespread in southern Tunisia, where it has been empirically used for its numerous therapeutic virtues against the rheumatism. It is also used sometimes like condiment, replacing onion (Cuénod, 1954). Contrarily to the cultivated garlic (*Allium sativum* L.) that makes the object of several studies treating its morphological, agronomic and molecular aspects (Baghalian *et al.*, 2006, 2005; Senula and Keller, 2005; Maab and Klaas, 1995)

the studies that are interested to the rosy garlic (*Allium roseum* L.) are so limited. However these studies are dedicated mainly to the morpho phenologies (Jendoubi *et al.*, 2001) and cyto-caryologic aspects (Ferchichi, 1997).

The selection of a suitable plant material and the propagation of its culture constitute, in fact, the first step to relieve these natural populations. The present survey has been dedicated to characterize the reproductive potentialities of several accessions of *Allium roseum* L. from different bioclimatic stages in Tunisia.

This research appears in the setting of an approach of valorization of this species and it aims in second stage to select the effective genotype which can be cultivated to big scale. Since 2001, agronomic and morphological characterization of Tunisian *Allium roseum* material has been conducted in southern Tunisian area where the reference collection of this specie was planted, near Arid Region Institute (IRA). The morphological descriptors used concern the reproductive system and the intra-specific variability in southern Tunisian accessions. This article describes the results of a study on the phenotypic variability of this wild species in areas of its distribution in all the Tunisia using certain reproductive vigor descriptors, with a view to validating, or not, various natural populations defined during surveys.

MATERIALS AND METHODS

Planting material: The planting material studied is composed of 28 wild Tunisian populations of *Allium roseum* L. In this study, the term 'population' is used for plants, constituted in the same level of ploidy, they have the same reproductive system and derived from same habitat, whose size is defined by distances of efficient inter-pollination (Pernès, 1984).

Each wild population was thus represented in the study by 20. The *Allium roseum* plants originated from 5 bioclimatic areas from along the country. For subsequent analyses, these populations were coded from 1 to 28 (Table 1, Fig. 1).

Measurements: A preliminary study was carried out in 2006 during the main finished seeds maturity when the species curls its vegetative cycle (between April and Mai). The descriptors tested were exclusively quantitative: Number of Seeds produced per plants (NS), Number of Small Bulbs (NSB), Number of Mean Bulbs (NMB), Number of Big Bulbs (NBB), Total Number of Bulbs (TNB), Seeds Weight (SW) and Weight of Total

Bulbs (WTB). Bulbs produced by plant are subdivided according to their diameter in three categories: They are qualified big (NBB), if the diameter is superior to 7 cm, means (NMB), if the diameter is understood between 4 and 7 cm and small (NSB) if the diameter doesn't pass 4 cm (Fig. 2).

Table 1: Bioclimatic regions distribution of *Allium roseum* accession's studied

Accession	Label	Bioclimatic stage ¹	Latitude	Longitude
Lemaya	1	AFW	35.0271	10.1626
Touila	2	AFW	35.0074	9.4051
Bir Ali	3	ASW	34.6303	10.1218
Bouhlel	4	SASW	35.3618	10.6695
Dokhane	5	SASW	34.9817	10.7463
Nasrallah	6	AFW	35.4357	10.8759
El Fjèe	7	ASW	33.5000	10.6421
Sidi Lefi	8	AFW	9.7940	35.1626
El Baten	9	AFW	9.9962	35.6994
Chgaig Elkhaoui	10	AFW	10.8899	32.6428
Elgrine	11	ASW	10.5530	33.6201
Dahar	12	AFW	10.0112	33.1332
Gsarghilene	13	S	9.7253	33.0222
Bir amir	14	S	9.9746	32.5871
Sidi Elagueb	15	S	9.8225	32.5368
Kamour	16	S	9.6563	32.5368
Oued Dkouk	17	AFW	10.3008	32.6050
Skhira	18	ASW	10.1216	34.3598
Borj Bourguiba	19	S	9.9230	32.2028
Tiert	20	S	10.1570	30.9683
Mechehed elmark	21	S	10.1420	31.6413
Elmenzla	22	S	10.5058	31.7721
Tamegza	23	AFW	8.1166	34.4675
Elfrid	24	AFW	8.0726	34.4835
Le kef	25	SACW	8.4838	35.9885
Samaillette	26	ASW	10.9240	33.2875
Henchir snoussi	27	ASW	9.8423	34.1370
Haddej	28	AFW	9.4525	34.4050

Bioclimatic stages are defined according to pluviothermic coefficient of Emberger (1966) $Q^2 = 2000 \times P/M2-m$ where: P is the yearly average in mm of the pluviometry, M is the average of the most elevated temperatures of the hottest month (°k) and m is the average of the lowest temperatures of the coldest month (°k). ASW: Arid at Soft Winter, SASW: Semi-Arid at Soft Winter, ASW: Arid at Soft Winter, SACW: Semi-Arid at Cool Winter and S. Saharian

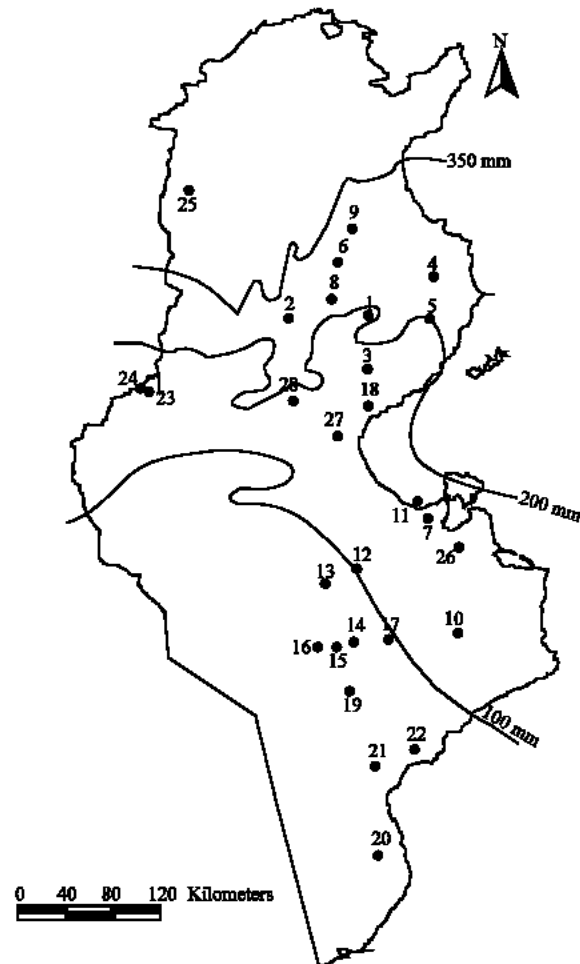


Fig. 1: Bioclimatic regions distribution of studied *Allium roseum*'s accession



Fig. 2: Genetic collection of *Allium roseum* installed in Arid Area Institute parcel

Statistical methods: An initial analysis of variance including the factor population was carried out. The data of the 7 variables were first analysed separately, then by a multivariate analysis of variance. This method is a generalization of the univariate analysis and uses the same model, but takes into account simultaneously the set of all variables along with their correlations. It is therefore more powerful for the detection of significant effects (Lachenaud *et al.*, 1999). The null hypotheses are controlled with the F test used also in the univariate analysis of variance (Rao, 1973). The multiple comparisons of populations' means were carried out separately on the 7 variables using the Duncan test. Discriminant analysis was used for comparison and graphic representation of the different populations. This multivariate analysis method consists in searching linear combinations of the original variables enabling the best possible separation of the populations (Lachenaud *et al.*, 1999). These new variables, or discriminant variables, correspond to the discriminant axes of the graphic representation (Saporta, 1990). The distance associated with discriminant analysis is the Mahalanobis distance. It was used to calculate the distances between the population means (Hebert and Vincourt, 1985) and the nullity of each distance was tested separately. The analyses of variance were carried out using the GLM procedure of the SAS software (SAS, 1990) and the discriminant analyses with the CANDISC procedure.

RESULTS

Preliminary results: Univariate and multivariate analysis of the elementary data relative to the Tunisian material in accordance with the described model and for the seven descriptors, did not reveal any bioclimatic stages effect on the population means, or any population bioclimatic stages interaction. That meant that observations for all bioclimatic stages could be grouped to analyse the population effect (Table 2).

Correlations between descriptors: A study of correlations, involving the entire data set for the Tunisian material, revealed that the majority of seven descriptors were significantly correlated, though with very heterogeneous values for the coefficients of determination R^2 (between 10^{-8} to 0.998) ones, thereby vindicating a separate study of the descriptors (Table 3).

Revealing variability

'Population and Bioclimatic stages' effect: An analysis of variance was carried out on the data for all Tunisian material: the 'population' effect was highly significant for the seven descriptors, with critical levels of 0.0001 for all studied descriptors (except this related to weight seeds). The Duncan test at 5% revealed one to ten groups of means depending on the descriptors (Table 4). The values of the controls could be compared to those of Jendoubi *et al.* (2001). It was found that the majority of populations always figured in the leading group, whereas populations 1, 2, 4 and 6 were always in the last group. For the 'number of small bulbs' descriptor, populations 1, 2, 4 and 6 stood out from the others. The relative amplitude of variation in the means of the study.

When considering the amplitude of variation according to bioclimatic stages values compared to the general mean, values of 46.21, 5.41, 20.32, 0.73, 24.85 and 0.15% were obtained, respectively.

Populations compared to the general mean was:

- 58.78% for, Number of Seeds produced per plant (NS)
- 5.4% for, Number of Small Bulbs (NSB)
- 12.29% for, Number of Mean Bulbs (NMB)
- 2.55% for Number of Big Bulbs (NBB)
- 5.27% for Total Number of Small Bulbs (TNB)
- 3.70% for Seeds Weight (SW)
- 1.89% for Weight of Total Bulbs (WTB)

Discriminant analysis and mahalanobis distances: A discriminant analysis was carried out on all the data relative to the Tunisian *Allium roseum* L. Collection

Table 2: Analysis of variance (pop bioclimatic stages, pop. x bioclimatic stages model), 28 populations studied during seeds maturity periods. (MS = Mean Square, **: significant at 1%)

Source	df	NS		NSB		NMB		NBB	
		MS	F	MS	F	MS	F	MS	F
Population	27	4.05 (10 ⁶)	4.93 (*)	2.65 (10 ³)	18.31 (**)	4.21 (10 ⁶)	21.04 (**)	41.49	4.17 (*)
Bioclimatic stage (BS)	4	4872.17	0.594	379.87	2.61	5238.93	2.01	20.736	2.61
Pop×BS	23	1340.5	0.163	279.13	1.92	3649.16	1.82	6.78	0.683

Table 2: Continued

Source	df	TNB		SW		WTB	
		MS	F	MS	F	MS	F
Population	27	6.93 (10 ⁶)	19.93 (**)	0.270	1.63	594.93	13.89 (**)
Bioclimatic stage (BS)	4	9141.78	2.62	0.193	1.166	37.49	0.876
Pop×BS	23	6355.32	1.82	0.05	0.305	21.48	0.502

(Table 5 and Fig. 3). The coefficients of canonical correlation r measure the discriminant power of each axis: $r = 1$ if all the individuals in the same group are projected onto the same point on the discriminant axis; $r = 0$ if group centre projections on the discriminant axis merge. The quantities $r^2/(1-r^2)$ represent the dispersion of group centers compared to within-group dispersion. Inertia (and cumulated inertia) corresponds to the raw (and cumulated) proportions of the previous quantities on each axis. The first two axes have a separating power superior to 89% of that of the seven initial variables. Axis 1, which is the most discriminant, was highly correlated with (in absolute value) Seeds Number (SN) and Seeds Weight (SW), whereas others descriptors (NSB, NMB, NBB and TNB) were correlated with axis 3. The Weight of Total Bulbs (WTB) was in an intermediate position, though it was less correlated with plane 1-2. The first bisector (Fig. 3a and b) was a size factor and opposed groups with seeds number and bulbs number (for 3 categories of bulbs) (population 1) and groups with higher number of seeds and less number of bulbs (population 14).

It was possible to project the populations, according to their bioclimatic stages. Despite their high variability, they stood out completely from corresponding bioclimatic

stage. So it was possible to find two or more populations from semi arid at cool winter climate (population 25) regrouped with populations from at soft winter (population 28).

Tests of Mahalanobis distance nullity between populations were carried out separately for each distance. The critical level α of each test was fixed using the principle of the Sidak test such that the overall level of α' for all the tests did not exceed 5%. As the number of distances to be tested between the 28 populations was $28 \times (28-1)/2 = 378$, α was such that $(1 - \alpha)^{378} = 1 - \alpha'$. The Mahalanobis D^2 distances, with an

Table 3: Values of the Pearson coefficients of correlation R (and R^2 in brackets) (*: significant at 5%, **: significant at 1%)

	NS	NSB	NMB	NBB	TNB	SW
NSB	-0.395* (0.156)					
NMB	-0.393* (0.154)	0.984** (0.968)				
NBB	-0.057 (0.030)	0.744** (0.553)	0.812** (0.659)			
TNB	-0.384* (0.184)	0.987** (0.974)	0.999** (0.998)	0.815** (0.664)		
SW	0.795** (0.632)	-0.108 (0.011)	-0.156 (0.024)	-0.0001 (10 ⁻⁵)	-0.142 (0.020)	
WTB	-0.350 (0.122)	0.949** (0.900)	0.979** (0.958)	0.879** (0.772)	0.979** (0.958)	-0.150 (0.022)

Table 4: Mean values per population for the seven descriptors (in gramme for SW and TWB). The different non-significant values are grouped under the same letter (The control values are given for comparison)

	NS	NSB	NMB	NBB	TNB	SWT	WB
1 AF	31.33h,i,j	47.88a	219.28a	14.73a	281.88a	0.09a	29.96a
2 AF	55.54h,i,j	60.13a	214.17a	8.43b,c	282.74a	0.17a	25.84a
3 AD	95.24f,g,h,i,j	13.00b	65.70c	4.82b,c,d	83.52c	0.28a	10.23b,c
4 SAD	0.00j	40.29a	162.50 a,b	9.70 b	212.48 a,b	0.00 a	28.31 a
5 SAD	0.00j	13.54b	62.83 c	2.74 d	79.11 c	0.00 a	9.79 b,c
6 AF	48.19h,i,j	42.88a	138.34b	4.66c,d	185.88b	1.47a	17.13b
7 AD	77.50 g,h,i,j	6.31b	40.00c	3.72c,d	50.03c	0.25a	6.50c
8 AF	225.92 b,c,d,e,f	11.17b	51.25c	3.83c,d	66.25c	0.81a	7.86b,c
9 AF	140.68c,d,e,f,g,h,i,j	3.21b	16.16c	1.00d	20.37c	0.50a	2.54c
10 AF	123.25c,d,e,f,g,h,i,j	3.50b	14.50c	1.25d	19.25c	0.40a	2.02c
11 AD	107.31d,e,f,g,h,i,j	1.00b	6.00c	0.63d	7.63c	0.38a	1.05c
12 AF	0.00j	2.72b	9.20c	1.00d	12.92c	0.00a	1.59c
13 S	0.00j	2.82b	9.36c	1.45d	13.64c	0.00a	2.09c
14 S	127.82c,d,e,f,g,i,j	4.06b	8.65c	0.88d	13.59c	0.44a	1.47c
15 S	18.00i,j	2.36b	5.07c	0.86d	8.29c	0.05a	1.10c
16 S	241.50b,c,d,e	6.53b	11.34c	1.16d	19.03c	0.77a	2.02c
17 AF	211.50b,c,d,e,f,g	4.00b	15.57c	1.00d	20.57c	0.67a	2.16c
18 AD	254.83b,c	2.42b	8.00c	3.67c,d	14.08c	0.80a	2.51c
19 S	150.67c,d,f,g,h,i	1.00b	8.42c	0.92d	10.33c	0.51a	1.38c
20 S	292.33b	1.44b	7.78c	1.33d	10.56c	1.22a	1.59c
21 S	158.20b,c,d,e,f,g,h,i	1.00b	5.00c	0.80d	6.80c	0.48a	0.89c
22 S	93.57f,g,h,i	2.57b	7.29c	0.57d	10.43c	0.33a	1.29c
23 AF	73.96g,h,i,j	2.19b	3.48c	2.22d	7.89c	0.27a	1.88c
24 AF	414.10a	1.86b	13.14c	8.57b,c	23.57c	1.60a	6.59c
25 SAF	167.57	1.57b	7.00c	5.43b,c,d	14.00c	0.36a	4.33c
26 AD	104.71e,f,h,i,j	4.71b	9.41c	1.06d	15.18c	0.36a	1.61c
27 AD	98.92f,h,i,j	0.85b	6.31c	2.58d	9.73c	0.32a	1.94c
28 AF	246.62b,c,d	0.31b	6.62c	3.31d	10.23c	0.91a	2.40c
Means	127.16	10.19	40.44	3.30	53.93	0.48	6.36
SAD	0.00a	26.92a	112.67a	6.22a	145.80a	0.00a	19.05a
AF	142.83a	16.35a	63.79a	4.54a	84.69a	0.62a	9.08a, b
AD	123.09a	4.72a	22.57a	2.74a	30.03a	0.39a	3.97b
S	135.26a	2.72a	7.86a	0.99a	11.58a	0.47a	1.47b
SAF	167.57a	1.57a	7.000a	5.43a	14.00a	0.36a	4.33

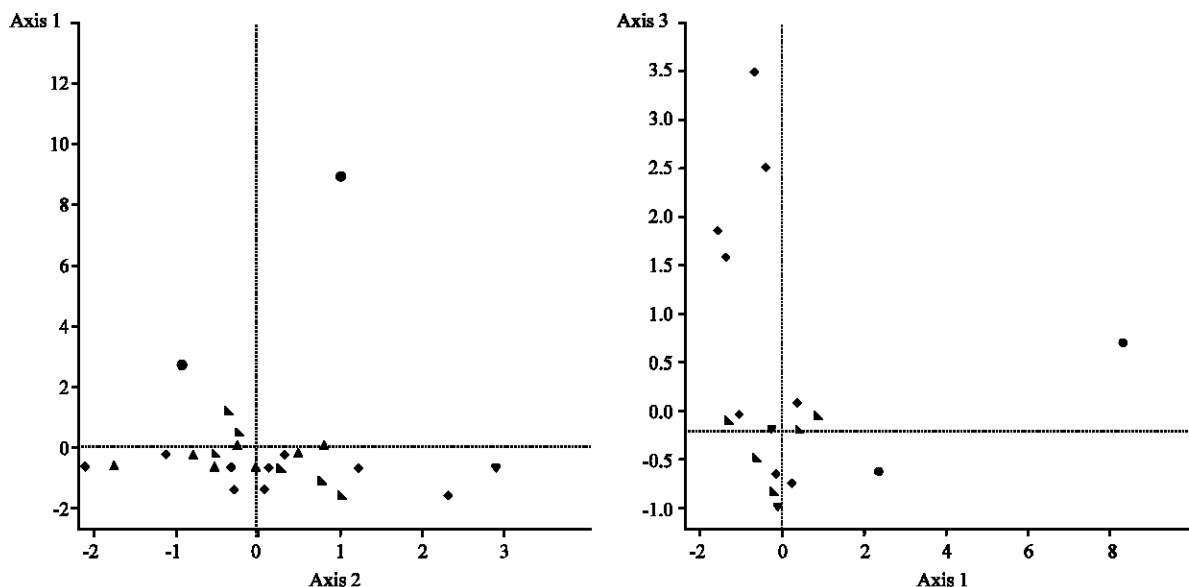


Fig. 3 a, b: Representation of the centers of gravity for Tunisian *Allium roseum* plants populations, in the planes of canonical axes 1 and 2, (2a) and planes canonical axes 1 and 3 (2b). In (2b), representation of the centers of gravity in the plane of axes 1 and 3. and indicate respectively: Semi-arid at soft winter, Saharian, Arid at cool winter, Arid at soft winter and Semi-arid at cool winter bioclimatic stages

Table 5: Factorial discriminant analysis parameters, where r = canonical correlation

Axis	r ²	r ² /1-r ²	Inertia	Between-group correlation with						
				SN	NSB	NMB	NBB	TNB	SW	TWB
1	0.519	1.080	0.789	-0.976	0.549	0.587	0.425	0.577	-0.846	0.699
2	0.405	0.682	0.104	0.127	-0.210	-0.179	0.284	-0.167	-0.244	-0.046
3	0.237	0.312	0.090	0.07	0.788	0.803	0.856	0.798	0.415	0.712

Table 6: Mahalanobis distance (squared) between the different populations. The significantly non-null distances are in bold face

Pop	1	2	3	4	5	6	7	8	9	10	11	12	13	14
2	3.86													
3	3.87	5.08												
4	6.95	11.82	6.03											
5	4.79	4.84	0.62	5.18										
6	5.09	1.59	3.00	9.61	2.46									
7	4.75	5.80	0.12	7.40	0.73	3.32								
8	6.10	6.19	0.75	8.91	2.28	3.92	0.88							
9	7.24	6.15	0.72	9.84	1.42	3.32	0.48	0.52						
10	7.16	6.04	0.81	10.47	1.54	2.98	0.47	0.73	0.05					
11	7.51	6.40	0.84	18.21	1.37	5.11	0.46	0.90	0.06	0.02				
12	7.00	6.26	1.09	23.64	0.90	6.39	0.59	2.24	0.77	0.58	0.44			
13	6.82	6.64	0.94	11.64	0.81	4.03	0.48	2.16	0.80	0.64	0.49	0.03		
14	7.81	6.45	0.97	19.60	1.65	5.39	0.61	0.78	0.06	0.03	0.04	0.64	0.64	
15	7.30	6.47	1.06	15.64	1.01	4.61	0.56	2.00	0.61	0.44	0.31	0.01	0.04	0.46
16	9.39	7.72	1.84	38.85	3.34	11.57	1.64	0.49	0.51	0.63	0.77	2.29	2.32	0.51
17	8.31	6.74	1.34	18.38	2.61	5.56	1.14	0.36	0.21	0.30	0.42	1.71	1.78	0.28
18	8.71	9.64	1.91	16.87	4.17	8.10	1.67	0.62	1.13	1.18	1.39	2.95	2.80	1.14
19	7.63	6.60	0.89	14.95	1.78	4.57	0.58	0.55	0.02	0.04	0.07	0.86	0.90	0.05
20	9.82	8.79	2.27	14.16	4.25	5.62	2.13	0.51	0.93	1.14	1.33	3.27	3.27	1.06
21	8.01	6.84	1.05	7.24	2.02	2.40	0.70	0.58	0.06	0.06	0.10	0.96	1.00	0.05
22	7.55	6.28	0.87	8.91	1.25	2.64	0.48	1.05	0.10	0.06	0.01	0.34	0.39	0.05
23	7.13	7.60	0.75	23.60	1.37	8.67	0.33	1.24	0.45	0.35	0.29	0.41	0.29	0.33
24	13.78	19.47	7.20	32.59	11.46	27.64	7.34	4.83	7.31	7.66	8.04	10.75	10.11	7.53
25	7.79	11.32	1.66	7.82	3.56	6.32	1.45	1.54	1.94	1.99	2.05	2.83	2.42	1.94
26	7.59	6.34	0.92	19.24	1.48	5.20	0.53	0.96	0.11	0.04	0.03	0.44	0.47	0.02
27	6.80	7.45	0.69	24.42	1.61	8.70	0.28	0.96	0.36	0.26	0.25	0.57	0.48	0.29
28	8.42	9.46	1.64	17.01	3.75	8.13	1.43	0.48	0.95	1.04	1.21	2.72	2.56	1.01

Table 6: Continued

Pop	15	16	17	18	19	20	21	22	23	24	25	26	27
16	1.95												
17	1.44	0.11											
18	2.58	0.52	0.70										
19	0.68	0.42	0.15	0.92									
20	2.88	0.18	0.31	0.44	0.79								
21	0.75	0.33	0.12	0.84	0.01	0.72							
22	0.22	0.88	0.54	1.59	0.14	1.54	0.17						
23	0.29	1.29	1.03	1.35	0.45	1.94	0.49	0.27					
24	10.13	5.44	6.26	2.99	6.96	4.42	6.84	8.45	7.04				
25	2.57	2.02	2.13	0.82	1.83	2.15	1.83	2.16	1.14	2.90			
26	0.2926	0.73	0.46	1.34	0.12	1.37	0.13	0.02	0.24	7.94	1.96		
27	0.44	1.0727	0.79	0.99	0.32	1.57	0.35	0.29	0.05	6.43	1.01	0.24	
28	2.37	0.50	0.6128	0.03	0.78	0.38	0.73	1.41	1.20	3.12	0.77	1.21	0.86

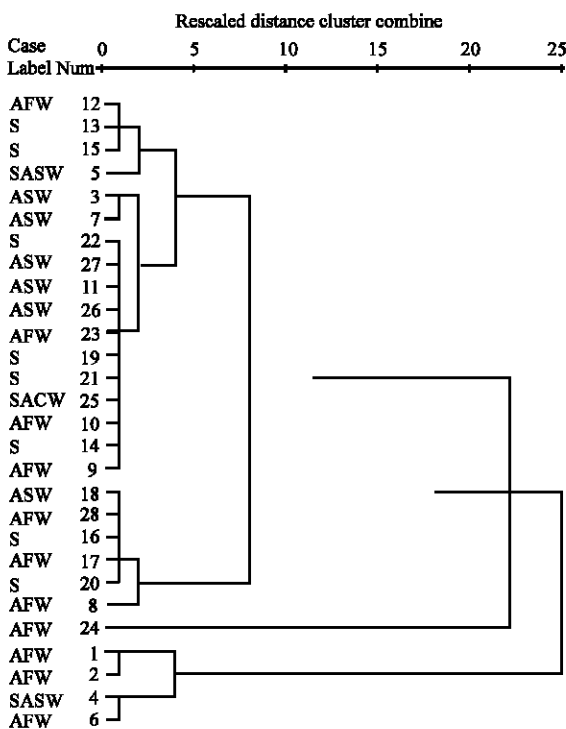


Fig. 4: Dendrogram of 28 populations of *Allium roseum* constructed with Euclidean formula, genetic distance matrix estimated from vigor reproductive descriptors data and clustered with the groups linkage methods

indication of those that are significantly non-null, are given in Table 6. Two great groups were clearly opposed: Populations 1, 2, 4, 6 and 24 on the one hand, the whole of others studied populations on the other hand. Although less marked, some opposition occurred within the second group: The distances separating populations 5, 20, 7 and 16, were significantly non-null. That's why the two enounced great groups can be divided into more homogenous subgroups.

The Mahalanobis D^2 distances can draw up a phylogenetic dendrogram relations revealed between studied populations (Fig. 4). We can find 4 groups which include many populations from different bioclimatic stages, the regrouped population could have near values for each studied descriptors.

DISCUSSION

By the end of the analyses, four main groups had been defined, one of which was itself divided into sub groups. The first comprised populations 12, 13, 15 and 5; The second comprised populations 3, 7, 22, 27, 11, 26, 19, 21, 25, 10, 14 and 9; The third group include populations 18, 28, 16, 17, 20 and 8 the latest group comprised populations 1, 2, 4, 6 and 24. This division confirmed the presence of more than bioclimatic stage in the some group (we found four bioclimatic stages in the second group). In addition this division confirmed contrast already seen between populations 1, 2, 4 and 6 on the one hand and the whole of other populations on other hand for certain reproductive vigor descriptors, including number. The discriminant analysis showed that the *Allium roseum*'s populations were very different from their bioclimatic originality.

The populations of same bioclimatic stage (e.g., semi-arid at cool winter) did not constitute a homogeneous set and short distances seemed to isolate certain populations, such as populations 23 and 24, which perhaps comprised related individuals, progenies of a few foundation plants. These two populations, which are 1000 m apart, were significantly separated. In fact, despite the short distance, they were not in communication nevertheless this species is qualified, like the majority of *Allium* subgenus species, an allogameous species (Jendoubi *et al.*, 2001; Ferchichi, 1997). The likeliest explanation is the coexistence of several varieties of *Allium roseum* L. in Tunisia. However Cuenod (1954) was confirmed that this species comprised more than four varieties, in fact '*Oderatissimum*' was the most rife one nevertheless in the southern part of the country (Ferchichi, 1997).

The populations 1, 2, 4, 6 and 24, proved to be genetically much closer to each other than any of the other populations and could constitute an original group. Moreover, the Mahalanobis distances were always significant (between populations 1, 2, 4, 6 and 24 and the whole of others populations).

As regards the *Allium roseum* plants, for which little is known about the last 2 aspects and whose effectiveness has perhaps been underestimated as for numerous other wild species, geographical remoteness could not still explain the genetic divergence of *Allium roseum* populations (Lachenaud *et al.*, 1999). The precise role played by the density of adult plants, in both isolated populations and throughout the zone, remains a major factor yet to be determined in order to explain the structuring of variability, but it could be slight (Doligez and Joly, 1997). The 'reproductive strategy' of *Allium roseum* L. is to encourage the scattering of the species by the vegetative way in the dependent to the sexual one or inversely. Indeed such hypothesis must be testified, nevertheless, until now no survey has been dedicated to study the floral biology, nor however for the resources allocation of the species. Such aspects would be in fact, with high utility in order to know the evolutionary tentatives within reproductive system of this species.

REFERENCES

- Baghalian, K., M. Reza Naghavi, A.Z. Seyed and N.B. Hassanali, 2005. Post-planting evaluation of morphological characters and allicin content in Iranian garlic (*Allium sativum* L.) ecotypes. *Scientia Horticult.*, 103: 155-166.
- Baghalian, K., M.R. Sanei, M.R. Naghavi, A. Khaligh. and H.A. Naghdi Badi, 2006. Post-culture evaluation of morphological divergence in Iranian garlic ecotypes. *Scientia Horticult.*, 107: 405-410.
- Cuénod, A., 1954. Flore de la Tunisie. Office de l'Expérimentation et de la Vulgarisation Agricole de Tunisie, pp: 287.
- Doligez, A. and H. Joly, 1997. Genetic diversity and spatial structure within a natural stand of a tropical forest tree species, *Carapa procera* (Meliaceae). In: French Guiana. *Heredity*, 79: 72-82.
- Emberger, L., 1966. Une classification biogéographique des climats. *Recherches et travaux des laboratoires de Géologie, Botanique et Zoologie. Faculté des Sciences Montpellier France*, 7: 1-43.
- Ferchichi, A., 1997. Contribution à l'étude caryologique, caryosystématique, morpho-biologique et écologique de la flore de la Tunisie présaharienne. Thèse d'Etat en Sciences biologiques. Uni. Tunis II., pp: 214.
- Hébert, Y. and P. Vincourt, 1985. Mesures de la Divergence Génétique. Deux Distances Calculées Sur des Critères Biométriques. In: *Les Distances Génétiques, Estimations et Applications*, M. Lefort Buson et D. De Vienne, (Eds.), INRA (Paris).
- Jendoubi, R., M. Neffati, B. Henchi and A. Yobi, 2001. Système de reproduction et variabilité morpho-phénologique chez *Allium roseum* L. *Plant Genetic Resou.*, 127: 29-34.
- Lachenaud, P., F. Bonnot and G. Oliver, 1999. Use of floral descriptors to study variability in wild cocoa trees (*Theobroma cacao* L.) in French Guiana. *Gene. Resou. Crop Evolu.*, 46: 491-500.
- Maab, H. and M. Klaas., 1995. Intraspecific differentiation of garlic (*Allium sativum* L.) by isozyme and DAPD markers. *Applied Genet.*, 97: 89-97.
- Marcucci, R. and N. Tornadore, 1997. Intraspecific variation of *Allium roseum* L. (Alliaceae). *Webbia*, 52: 137-154.
- Pernès, J., 1984. Gestion des Ressources Génétiques des Plantes. Tome II, (Eds.), Lavoisier Tec et Doc. Paris, pp: 384.
- Rao, C.R., 1973. Linear statistical inference and its applications (2nd Edn.), John Wiley and Sons, Inc. New York.
- Ricroch, A., R. Yockteng, S.C. Brown and S. Nadot, 2005. Evolution of genome size across some cultivated *Allium* sp. *Genome*, 48: 511-520.
- Saporta, G., 1990. Probabilités, analyse des données et statistique. Technip, Paris.
- Senula, A. and E.R.J. Keller, 2005. Diversity in a clonally propagated crop: Morphological characters in garlic compared with existing molecular classifications. Institut für Pflanzengenetik und Kulturpflanzenforschung Gatersleben (IPK) Corrensstr. 3 D-06466 Gatersleben, Germany.
- Stearn, W.T., 1992. How many species of *Allium* are known? *Kew. Mag.*, 9: 180-181.