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# Preliminary selection of *Lolium perenne* L. natural populations for pasture improvement purpose in rainfed Mediterranean conditions

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**Abstract.** Previous researchs have established how *Lolium perenne* L. genetic conservation, evaluation and development have priority for Mediterranean permanent pasture sustainability, and how new persistent and drought tolerant varieties could contribute to mixtures of annual and perennial legumes for pasture improvement purpose. Nevertheless, scarce is the availability of perennial ryegrass cultivars responding to semi-arid climates adaptation criteria; their persistence being reduced by incidences of summer droughts. The attempt to extend this availability has been started through a collection of germplasm and consequent field evaluation of natural populations, originating from different pedo-climatic sites of Corsica and Sardinia islands in the Mediterranean Sea, in terms of productive parameters, habit, phenology and persistence. The field evaluation trial was carried out at the Ottava experimental station of the Sassari University (570 mm average annual rainfall, North Sardinia, Italy). Starting from initial 30 natural populations, 11 preliminarily selected accessions were compared over three years, under rainfed conditions, following the UPOV rules for the identification of new cultivars, with the purpose to estimate the potential inclusion of most interesting accessions in programs that lead to varietal certification.

**Keywords.** *Lolium perenne* – Rainfed conditions – Semi-arid climate – Persistence.

## **Sélection préliminaire des populations naturelles de *Lolium perenne* L. dans le but de amélioration des pâturages dans les conditions non irriguées méditerranéens**

**Résumé.** Recherches antérieures ont établi comment la conservation génétique, l'évaluation et le développement des graminées pérennes, ont la priorité pour la durabilité des pâturages permanents méditerranéens, et comment de nouvelles variétés, persistantes et tolérantes à la sécheresse, pourraient contribuer aux mélanges de légumineuses annuelles et pérennes pour objectif d'amélioration des pâturages. Cependant, la disponibilité de cultivars de ray-grass pérennes répondant à des critères de l'adaptation au climat semi-aride est rare, leur persistance étant réduite par l'incidence des sécheresses estivales. La tentative d'étendre cette disponibilité a été démarré à travers une collection de matériel génétique et par l'évaluation morpho-agronomique des populations natives, provenant de différents sites pédo-climatiques sur les îles de Corse et de Sardaigne dans la mer Méditerranée, en termes de paramètres productives, de phénologie et de persistance. L'étude de évaluation sur le terrain a été réalisée à la station expérimentale de Ottava de l'Université de Sassari (570 mm précipitations annuelles moyennes, du Nord Sardaigne, Italie). A partir de 30 initiales populations naturelles, 11 accessions persistantes ont été comparés sur trois ans, dans des conditions non irriguées, suivant les règles de l'UPOV pour l'identification de nouveaux cultivars, dans le but d'estimer l'inclusion éventuelle de des accessions plus intéressantes dans les programmes qui mènent à la certification variétale.

**Mots-clés.** *Lolium perenne* – Conditions non irriguées – Climat semi-aride – Persistence.

## **I – Introduction**

The maintenance of ecosystem services is becoming one of the most important component of European farm income, through the subsidies introduced by the European Union's common agri-

cultural policy (Lelièvre and Volaire, 2009); in this new contest, it is clear that semi-permanent improved pastures based on multifunctional mixtures of annual and perennial species may have a positive role (Franca *et al.*, 2007). In recent years, there have been advances in the availability of annual pasture species in the Mediterranean seed market, while progress remains to be made in selecting perennials useful for this purpose. Commercial perennial grasses, and even drought tolerant perennial ryegrass varieties, often do not persist, being not able to survive to hot and dry summer in Mediterranean environment (Franca *et al.*, 1995). Through the years, various germplasm collection campaigns were carried out in the Mediterranean Basin. Regional gene-pools of perennial ryegrass were established in France (Charmet and Balfourier, 1990), a core germplasm collection was carried out in Tunisia (Chakroun *et al.*, 1995) and a germplasm collection for breeding purpose was concluded in Italy (Falcinelli *et al.*, 1988). But, nevertheless, no selection process was completed and it is still difficult to find a perennial ryegrass variety able to compose a balanced and persistent multifunctional mixture for pasture improvement purpose. So, research programs should ensure a diversification of available perennial grasses germplasm for phenology, drought tolerance, summer dormancy levels, and dehydration tolerance.

The attempt to extend this availability has been started through a collection of germplasm and consequent field evaluation of natural populations, originating from different pedo-climatic sites of the islands of Corsica and Sardinia in the Mediterranean Sea, in terms of productive parameters, habit, phenology and persistence. In this paper, coverage values at the autumn establishment were used for observing the persistence capability of the populations, while data on plant morphology and productivity were utilized for clustering the persisting populations for similarity.

## II – Materials and methods

The 30 populations used in this experiment were sourced from a collection carried out in 2004 within the Vegetatio Interreg III A programme by ISPAAM (Sardinia, Italy) and ODARC (Corsica, France). On autumn 2005, collected seeds were sown in a experimental field in Sassari (Italy), 80 m a.s.l., average annual rainfall 547 mm, average annual temperature 16,2°C, on a calcareous sandy-lime soil. Seeds were sown in rows, 20 cm distant. Plots (1 m x 3 m) were arranged in a randomized block design with three replications.

The persistence of each population from autumn 2005 to spring 2008 was determined visually estimating the coverage percentage of the population in autumn. Only 11 populations re-established for three years and were included in the analysis. Also, with the aim of studying the morphological variability of the persisting populations, the following morphological characters were observed: tiller length, flag leaf length and width, spike length; number of sterile and reproductive tillers per plant, number of spikelet per spike, plant habit. In terms of productivity, dry matter yield and potential seed yield of the persisting populations were determined. Cluster analysis was performed on average values of morphological and productive data per population, only using data of the second year. Statgraphics Centurion (StatPoint Inc.) multivariate procedures (Squared Euclidean distances and Nearest Neighbour Method) were performed.

## III – Results and discussion

Data are referred to the 11 of the 30 collected populations that persisted for three years in the field in dry conditions. Cantoniera Atzeni is the only Sardinian population that persisted, the other 10 populations are original of Cap Corse (Corsica, France) (Fig. 1). All the populations improved their coverage in the second year, compared with the sowing year, and seven populations have followed the same trend even in the third year, reaching in most of the cases coverage percentages higher than the 50%. Only two populations, RG17 and RG6, decreased their re-establish-

Region	2006	2007	2008
cant.atzeni	46	62	71
RG1	21	62	54
RG2	29	62	46
RG6	21	38	21
RG7	29	38	46
RG9	12	62	71
RG11	46	54	87
RG15	46	54	62
RG16	29	62	71
RG17	29	38	21
RG18	38	54	62

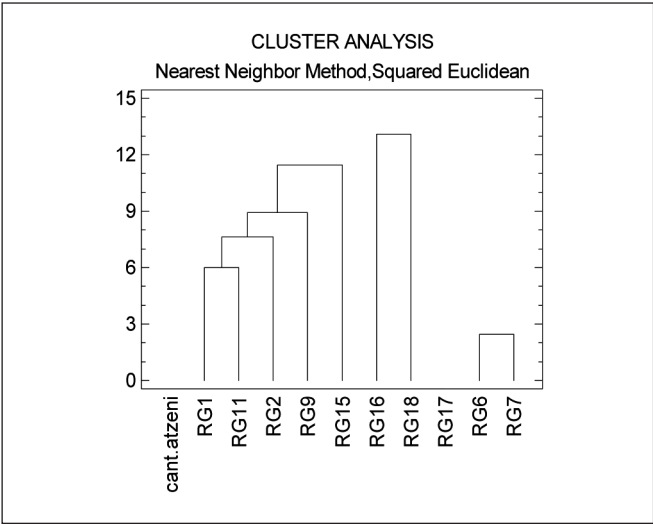
The correlation matrix (Table 1) shows a positive correlation between seed yield and dry matter yield, in opposition to results of previous breeders' studies on Mediterranean perennial grasses (Falcinelli *et al.*, 1988). Seed and dry matter yield are also significantly correlated with the number of reproductive tillers. Also, higher the plant height, higher the size of the vegetative traits, like tiller length and flag leaf length and higher the number of spikelets spike<sup>-1</sup>. Tall plants showed also higher number of reproductive tillers.

TL	TL					TL	Tiller length	
SL	0.893 ***	SL				SL	Spike length	
FL	0.872 ***	0.749 **	FL			FL	Flag leaf length	
SPT	0.814 **	0.656 *	0.886 ***	SPT		SPT	Spikelets spike <sup>-1</sup>	
RT	0.591 n.s.	0.453 n.s.	0.585 n.s.	0.504 n.s.	RT	RT	Reproductive tillers	
TT	0.541 n.s.	0.319 n.s.	0.632 *	0.571 n.s.	0.908 ***	TT	Sterile tillers	
PH	0.920 ***	0.709 *	0.892 ***	0.793 **	0.714 *	0.731 *	PH	
SEED	0.612 *	0.441 n.s.	0.370 n.s.	0.373 n.s.	0.780 **	0.665 *	0.590 n.s.	SEED
DMY	0.739 **	0.502 n.s.	0.721 *	0.584 n.s.	0.908 ***	0.880 ***	0.885 ***	0.765 **

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Cluster Analysis (Fig. 2) showed that Cantoniera Atzeni and RG17 had peculiar traits that made these two populations dissimilar to the others. In Table 2, the average values of the observed morphological and productive traits which characterize each cluster are reported.

The populations grouped in clusters 1 to 3 are more productive for seed yield and DMY. Cantoniera Atzeni is characterized by the elongation of the tiller and the spike, the erect habit and the early flowering. Cluster 2 groups five populations which differ from Cantoniera Atzeni mainly for shorter tiller and spike. Cluster 3 is composed by two populations (RG16 and 18), which are characterized by an high incidence of sterile tillers on the total number of tillers per plant. RG17 is constituted by very short plants, producing the lowest DMY. Cluster 5 is composed by two populations having short plants, but with a good tillering and late flowering.



**Fig. 2. Cluster Analysis of the 11 persisting populations for the main observed morphological and productive characters in the second year of the experimentation.**

**Table 2. Average values of the observed traits characterizing each cluster resulted from multivariate analysis**

Cluster	Population	TL cm	SL cm	FL cm	SPT nr.	RT nr.	TT nr.	PH cm	SEED g m <sup>-2</sup>	DMY g m <sup>-2</sup>	FLOWERING
1	CANT.ATZENI	61	39	17	29	234	291	39	24	1032	EARLY
2	RG 1-2-9-11-15	51	27	14	28	212	289	32	25	984	MID
3	RG 16-18	41	22	12	25	236	327	28	20	1043	MID-LATE
4	RG 17	34	23	7	21	130	146	8	15	179	MID-LATE
5	RG 6 - 7	35	20	11	25	133	227	14	10	284	LATE

## IV – Conclusions

Between the 11 persisting populations, a great variability of the morphological and productive characters was observed. The preliminary results of this research permits to show that is possible to develop a valorization program of different morphotypes of perennial ryegrasses, persistent in semi-arid rainfed conditions. Cantoniera Atzeni will represent the basic genotype for selecting a productive, erect and early flowering perennial ryegrass to be used for forage/hay production. Populations from Corsica RG 6 and 7 will be multiplied to be used in multifunctional mixtures with annual and perennial grasses and legumes, with cover crop purposes.

An investigation is being carried out currently on these 11 persistent populations of *L. perenne* for studying physiological drought resistance mechanisms on leaves and root, phenologic adaptations.

In conclusion, the results of this trial are preliminary and preparatory to the development of a research aimed at identification of new *Lolium perenne* varieties, persisting to Mediterranean semi-arid rainfed conditions and suitable for forage/hay production rather than for cover crops management.

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