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*in*

**Porqueddu C. (ed.), Ríos S. (ed.).**
**The contributions of grasslands to the conservation of Mediterranean biodiversity**

**Zaragoza : CIHEAM / CIBIO / FAO / SEEP**
**Options Méditerranéennes : Série A. Séminaires Méditerranéens; n. 92**

**2010**
pages 251-255

# Characterization of Moroccan populations of *Trifolium* and *Lotus* species

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**Abstract.** Annual forage legumes are an important resource for pasture improvement. They may be used to reduce erosion and improve grazing on marginal areas. In this study, we report morphological and biometrical evaluation of 70 populations belonging to 11 species of *Trifolium* and *Lotus* collected in different Moroccan regions. Plant height, leaf size, flowering time, flowering length, number of inflorescences per plant, seeds per inflorescence, 1000 seeds weight, seed plant production and dry matter yields were determined. High variability has been shown among and within species, especially for dry matter yield and flowering time. The morphological and agronomic variables were used to group studied populations by using a principal components analysis. Populations of *T. isthmocarpum* and *L. corniculatus* produced the highest biomass.

**Keywords.** *Trifolium* sp. – *Lotus* sp. – Wild collection – Multivariate analysis – Morocco.

***Caractérisation des populations marocaines de* Trifolium *sp. et* Lotus *sp.***

***Résumé.*** *Les légumineuses fourragères sont une ressource importante pour l'amélioration des pâturages. Elles peuvent être utilisées pour la prévention de l'érosion du sol et pour l'amélioration des zones marginales. Une étude morphologique et biométrique a été réalisée sur 70 populations appartenant à 11 espèces de* Trifolium *et* Lotus *collectées dans différentes régions du Maroc. La hauteur de la plante, la taille des feuilles, la date de floraison, la durée de floraison, le nombre d'inflorescences par plante, le nombre de graines par inflorescence, le poids de mille grains, la production de semences par plante et le rendement en matière sèche ont été déterminés. Une importante variabilité intra et inter-spécifique a été révélée, principalement pour le rendement en matière sèche et la date de floraison. Une analyse en composantes principales a été réalisée sur les caractères morphologiques et agronomiques.* T. isthmocarpum *et* L. corniculatus *avaient les rendements en biomasse les plus élevés.*

***Mots-clés.*** Trifolium *sp. – Lotus sp. – Collection sauvage – Analyses multivariées – Maroc.*

## I – Introduction

As a part of the Mediterranean centre of diversity, Morocco hosts many interesting crop and several wild species. Moreover, Morocco offers a great diversity of soil type, temperatures and rainfall. Native pastures have become rather deteriorated due to monoculture (cereals) and the intensive grazing. Many surveys have confirmed that pasture legumes are an important component of non-arable agriculture (Whitbread and Clem, 2006). In Morocco, pasture improvement is mainly concerned with subterranean clover (Amine and Baghati, 1997). However, subterranean clover has a limited adaptation to the various target regions, particularly the salt and drought affected areas. Indigenous populations can perform well or better than available cultivars in some environments. Therefore, the evaluation of wild ecotypes is important for selection of varieties adapted to different edaphic and climatic conditions in order to obtain higher productivity and to increase the pasture quality. *Trifolium* and *Lotus* are important forage legumes among the genera of the Fabaceae family (Garcia de los Santos *et al*., 2001; Ding *et al*., 2003). In this study we evaluate local populations of *Trifolium* and *Lotus* species for morphological and agronomic characteristics for possible exploitation and domestication.

## II – Materials and methods

The field experiment was conducted from October 2007 to August 2008, at the INRA's Guich experimental station, Rabat, Morocco (latitude 34°03′ N, longitude 06°46′ W, elevation 10.5 m) on a sandy soil, with a pH=7.3). The mean annual rainfall and temperature were: 350 mm, 22°C. The soil was fertilized with 45 kg/ha of $P_2O_5$ at sowing. The 70 populations used in this study were belonging to 11 species of genera of *Trifolium* and *Lotus*, collected at 12 Moroccan sites (Table 1). These species are: *T. arvense* L., *T. campestre* Schreb., *T. cherleri* L., *T. glomeratum* L., *T. isthmocarpum* Brot., *T. resupinatum* L., *T. scabrum* L., *T. stellatum* L., *T. tomentosum* L., *L. corniculatus* L. and *L. ornithopodioides* L. Seeds of each population were germinated in small pots filled with peat and maintained under greenhouse conditions. When the plants reached the fourth leaf stage, they were transplanted into the field. Each population was sown in a 1 m row of four plants and replicated twice. Row spacing was 0.2 m. The irrigation was performed through drip irrigation. The morphological characters were measured following the descriptor list of Andersen and Ellis Davies (1984). The measured variables were plant height (PH), leaf size (LS), flowering time recorded from germination at 50% of heading (FT); flowering length as days between the first and last date of flowering (FL), number of inflorescences per plant (IP), number of seeds per inflorescence (SI), seed plant production (P), 1000 seeds weight (TSW), dry matter yield g/m² (DM). The data were analyzed using the statistical software Genstat 5. Data analysis consisted of principal component analysis, using the morphological and agronomic characters to group the tested populations. Dry matter yield was subject to analysis of variance and means compared by Duncan's test.

**Table 1. Characteristics of collection sites of *Trifolium* and *Lotus* species in Morocco**

| Collection sites in Morocco | Site code | Mean annual rainfall (mm) | Soil pH | Latitude | Longitude | m† (C°) | M†† (C°) |
|---|---|---|---|---|---|---|---|
| Sidi Hajjaj | 1 | 242 | 8.8 | 33.13 | 7.36 | 6.3 | 29.8 |
| Sebt Brikyine | 2 | 200 | 6.7 | 32.26 | 8.06 | 4.3 | 39.4 |
| Oualidia | 3 | 294 | 8.3 | 32.6 | 9.01 | 7.9 | 29.1 |
| Sidi-Allal-Bahraoui | 4 | 399 | 6.0 | 34.04 | 6.51 | 4.8 | 31.6 |
| Dar-Bel-Amri | 5 | 400 | 6.1 | 34.21 | 6.44 | 4.8 | 31.6 |
| Sidi-Slimane | 6 | 320 | 8.0 | 34.26 | 5.89 | 4.3 | 36.5 |
| Moulay-Bousselham | 7 | 552 | 6.9 | 34.99 | 6.18 | 6 | 32 |
| Asilah | 8 | 614 | 7.8 | 35.61 | 5.69 | 7.8 | 27.8 |
| Marina smir (Tetouan) | 9 | 550 | 9.0 | 35.75 | 5.34 | 9. | 27.7 |
| Chefchaouen | 10 | 725 | 8.0 | 35.21 | 5.41 | 5.4 | 25.4 |
| Rommani | 11 | 410 | 7.3 | 33.77 | 6.62 | 5.3 | 36 |
| Maaziz | 12 | 390 | 7.0 | 33.71 | 6.14 | 5.3 | 36 |

†Mean minimum annual temperature.
††Mean maximum annual temerature.

## III – Results and discussion

### 1. Flowering time

The results of the days to flowering (Fig. 1), showed three major groups: (i) short cycle group (less than 110 days) represented by *L. corniculatus* (two populations) and *T. arvense*, *T. glomeratum*, *T. isthmocarpum*, *T. resupinatum*, *T. scabrum* and *T. tomentosum* (one population for each species); (ii) long cycle group (more than 150 days) for one population of *L.*

*ornithopodioides*, *T. stellatum* and *T. campestre*, and two *T. glomeratum* populations; and (iii) medium cycle group (110-150 days), represented by all the remaining species.

The flowering length is short (less than 20 days) for *T. cherleri*, *T. glomeratum*, *T. isthmocarpum*, *T. resupinatum* and *T. stellatum* and one *L. ornithopodioides* population. Conversely this length was longer than 40 days for *L. corniculatus* and *T. cherleri* (one population), and *T. Tomentoseum* (two populations).

## 2. Dry matter

Dry matter productivity differed greatly ($P < 0.001$) among species. *T. isthmocarpum* and *L. corniculatus* are the most dry matter producers (Fig. 2). *T. resupinatum* produced more than *L. ornithopodioides*, *T. glomeratum*, *T. tomentosum* and *T. cherleri*. *T. arvense*, *T. campestre* and *T. scabrum* have the similar low production. *T. stellatum* had the lowest production.

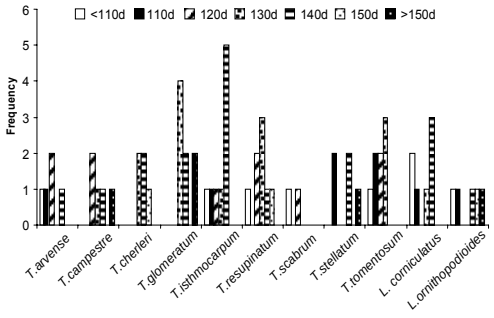


**Fig. 1. Frequency of *Trifolium* and *Lotus* populations according to the flowering time measured by days (d).**

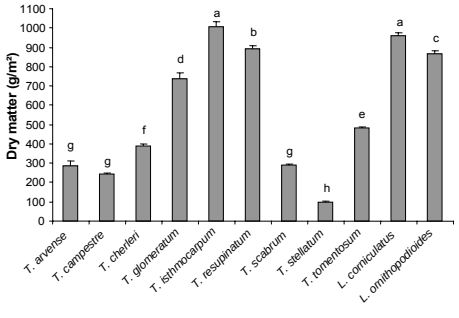


**Fig. 2. Dry matter yield (g/m²) of *Trifolium* and *Lotus* species. Means (±standard deviation) with the same letter are not significantly different (P ≤ 0.05).**

## 3. 1000 seed weight (TSW)

Statistical analyses of the TSW indicate the performance of *T. isthmocarpum,* especially those originated from Chefchaouen (2.45 g in average). This site is characterized by the highest annual rainfall and low temperatures. Abdelguerfi and Laouar (1997) cited that this species preferred the irrigated regions. The other species have shown less values of TSW: *L. corniculatus* (1.84 g), *T. cherleri* (1.15 g), *T. resupinatum* (1.07 g), *T. glomeratum* (0.69 g), *T. campestre* (0,68 g), *T. stellatum* (0.62 g), *T. tomentosum* (0.59 g) and *T. arvense*. *T. scabrum* was characterised by the lowest value (0.32 g).

## 4. Population groups

The morphological and agronomic variables measured (data partially not reported) of *Trifolium* and *Lotus* species were used for the principal component analysis (PCA). The first three PCA of clover species accounted for 75% of the variation within the data. Dry matter yield, plant height and leaf size were associated with the first component (PC1). While number of seeds per inflorescence, seed plant production, flowering time and flowering length were associated with the second component (PC2). The biplot of the first 2 principal components shows that populations from the regions with same climatic conditions are grouped, where the same species tend to pool together (Fig. 3a). For example, five populations of *T. isthmocarpum* (i2, i5, i7, i8 and i9) are grouped. They are characterized by high production, high plants, widest leaves

and presents a medium flowering time. The populations of this group are originated from Sidi-Allal-Bahraoui, Moulay Bousselham, Chefchaouen, Rommani and Maaziz. These areas are characterized by a high annual rainfall (between 400 mm and 730 mm) and a neutral pH soil. The five populations of *T. cherleri* are mainly characterized by high flowering length but low dry matter yield and a short height. The three populations of *T. scabrum* are early flowering, and are characterized by a very low level of 1000 seeds weight. Populations within the species *T. glomeratum* and *T. resupinatum* are characterized by high seed plant production and an important number of inflorescences per plant. The first three PCA of *Lotus* species accounted for 70% of the variation within the data. PC1 is associated with 1000 seeds weight, dry matter yield, seed plant production and plant height. PC2 is associated with flowering parameters (flowering time and flowering length), number of inflorescences per plant and leaf size. The graphic representation of PC1 and PC2 (Fig. 3b) shows that two populations of *L. corniculatus* (L3 and L4) and one population of *L. ornithopodioides* (o4) are the high productive species with a medium flowering time.

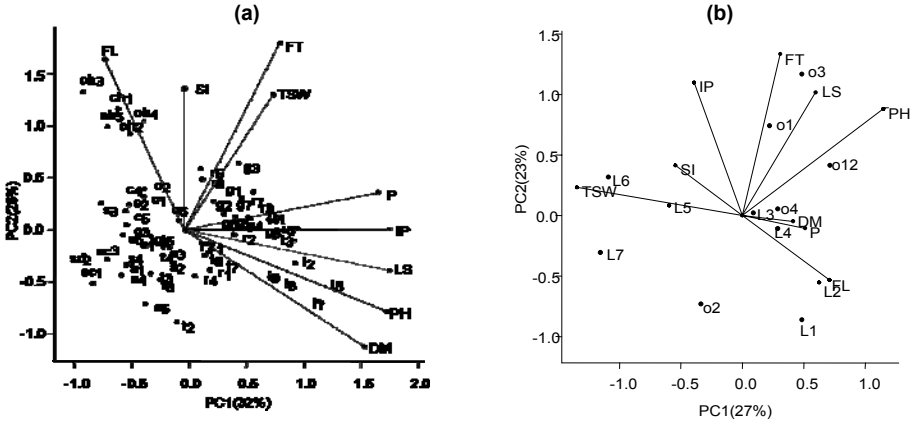


**Fig. 3. Biplot of the first 2 dimensions in the principal component analysis of variables among different populations. (a) *Trifolium* sp. (b) *Lotus* sp. (a: *T. arvense*; c: *T. campestre*; ch: *T. cherleri*; g: *T. glomeratum*; i: *T. isthmocarpum*; r: *T. resupinatum*; sc: *T. scabrum*; s: *T. stellatum*; t: *T. tomentosum*; L: *L. corniculatus*; o: *L. ornithopodioides*; 1 to 12: corresponding site).**

## IV – Conclusion

These preliminary results showed the existence of some variability concerning agronomical traits, particularly, flowering time and dry matter yield. Furthermore, the survival of most species seems to be linked to the environmental conditions such as rainfall and temperature. However, this study should be carried on by conducting further physiological evaluation on resistance to abiotic stresses. This is important to obtain more information on the impact of environmental factors on the geographic species and target more efficiently selection of varieties to be domesticated.

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