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Plant breeding

Genetic variability for morphology, growth and forage yield among perennial diploid and tetraploid lucerne populations (*Medicago sativa* L)

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Summary — The wide diversity within the *Medicago sativa* complex is well known, but comparative data on different populations representing the sub-species *sativa* and *falcata* are lacking. Morphological and agronomic characters of 25 populations or varieties of lucerne belonging to the *M sativa* complex were studied at Lusignan (France) in 1993 and 1994. The landraces and varieties of the sub-species *sativa* were clearly different from the wild populations of the 2 sub-species *sativa* and *falcata* by their erect growth habit, their fast regrowth after cutting, the absence of rhizomatous plants, their tall height and their high forage yield. Within the cultivated populations, those originating from northern France had a higher winter resistance than the Mediterranean populations, and higher yields. The winter resistance trait and the variegated flowers of the French *sativa* landraces show that they have integrated some traits belonging to sub-species *falcata*. The group of wild populations contained diploid and tetraploid populations, and *sativa* or *falcata* populations. Among the characters analysed, only flower colour made the distinction possible between the wild *sativa* populations from Spain (Mielga) with purple flowers and the *falcata* populations with yellow flowers. This convergence of forms between the 2 sub-species could be a strategy of tolerance to animal feeding and to treading.

Medicago sativa **L = lucerne / population / morphology / agronomic trait / genetic variability**

Résumé — Variabilité génétique pour la morphologie, la croissance et le rendement en matière sèche chez la luzerne pérenne diploïde et tétraploïde (*Medicago sativa* L). *La grande diversité du complexe* Medicago sativa *est bien connue, mais des données comparatives de différentes populations représentant les sous-espèces* sativa *et* falcata *manquent. Les caractéristiques morphologiques et agronomiques de 25 populations ou variétés de luzerne appartenant au complexe* M sativa *ont été étudiées à Lusignan (France) en 1993 et 1994. Les populations et variétés cultivées de la sous-espèce* sativa *se distinguent nettement des populations sauvages des 2 sous-espèces* sativa *et* falcata *par leur port dressé, leur repousse rapide après une coupe, leur hauteur élevée et leur fort rendement. Parmi les populations cultivées, celles originaires du nord de la France ont une résistance à l'hiver supérieure aux méditerranéennes, ainsi qu'un meilleur rendement. Par leurs caractères de résistance à l'hiver et leurs fleurs bigarrées, ces populations* sativa *françaises montrent qu'elles ont intégré certains caractères provenant de la ssp* falcata. *Le groupe des populations sauvages comprend à la fois des populations diploïdes et tétraploïdes,* falcata *ou* sativa. *Pour les caractères analysés, seule la couleur des fleurs permet de distinguer les populations sauvages* sativa *d'Espagne (Mielga) à fleurs violettes des populations* falcata *à fleurs jaunes. La convergence de forme de ces 2 sous-espèces pourrait représenter une stratégie de tolérance au pâturage.*

Medicago sativa ***L = luzerne / population / morphologie / caractère agronomique / variabilité génétique***

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INTRODUCTION

Perennial lucerne (*Medicago sativa* L) exists as a cultivated but also as a wild species. A large genetic variability is available within this species. Several authors have debated about taxonomic classification (Lesins and Lesins, 1979; Sinskaya, 1950, cited by Lesins and Lesins, 1979; Quiros and Bauchan, 1988). Quiros and Bauchan (1988) consider that the *M sativa* complex contains 2 main sub-species and 1 less widespread sub-species:

(1) Subspecies *sativa*, to which belong most of the cultivated varieties or landraces, is characterised by purple flowers, a tap root, an erect growth habit, coiled pods, a poor winter hardiness and no dormancy. The geographical origin of this sub-species is located in the Middle-East, and the zone of diversification is the Mediterranean area.

(2) Sub-species *falcata* is mainly wild, with yellow flowers, a prostate growth habit, fasciculate roots, sickle-shaped pods, strong winter hardiness and winter dormancy. It originates from Central Asia, and is diversified in northern Eurasia.

(3) Sub-species *glutinosa*, with yellow flowers, coiled and glandular-haired pods. It originates from Caucasia but is not widespread elsewhere.

Within each of these sub-species, genetic evolution occurred, which was mainly driven by 2 processes. Firstly, geographical diversification of lucerne was achieved by an adaptation to climate, soils or pests. Secondly, man has bred lucerne for its use and modified agronomically important traits. Furthermore, in Europe, both sub-species coexist and crosses have occurred for ages (Lesins and Lesins, 1979). In both ssp *sativa* and *falcata*, diploid and tetraploid forms exist; the tetraploids are usually more vigorous than diploids (Dunbier *et al,* 1975; Volenec, 1988) and are more frequently used by man for cultivation. The diploid form of *sativa* is called *coerulea*, and the diploid form of *falcata* is called *falcata*, *romanica* or *quasifalcata*. In Spain, wild forms of the ssp *sativa*, called Mielga, exist and are characterised by a prostate growth habit but purple flowers. In that country, lucerne was first introduced in Roman times (Bolton *et al,* 1972), and Mielga probably originated from these populations (Delgado Enguita, 1989). The cultivated Spanish populations were then introduced during the Moorish invasions in the 8th century (Hidalgo Maynar, 1966).

Another species related to the *M sativa* complex is *M glomerata* (Quiros and Bauchan, 1988). This originates from the Mediterranean Alps or North Africa and is intermediate between ssp *sativa* and ssp *falcata* because of its coiled pods and yellow flowers.

All the available populations (varieties, landraces or wild populations) can have specific features of interest for breeding, such as characters for adaptation, resistance to pests or stresses, and agronomic value. This study presents an evaluation of the genetic diversity among perennial *Medicago* for morphological, agronomic and growth traits. The relationships between the various populations and sub-species were investigated. A further paper will deal with characters of forage quality (digestibility), disease and nematode resistance and saponin countent.

MATERIAL AND METHODS

Twenty-five populations were studied, as described in table I. In the ssp *sativa*, 5 French landraces and varieties, 3 populations from Morocco (Birouk and Guy, 1986; Birouk *et al,* 1989), 2 landraces from Tunisia, 4 Spanish landraces and 3 Spanish wild Mielga, and 1 diploid *coerulea* population were chosen. In the ssp *falcata*, a Canadian diploid variety, 2 French wild tetraploid populations, an Ukrainian tetraploid population, and 2 Russian diploid populations called by their synonymous *romanica* and *quasifalcata* were studied. A *glomerata* population was included.

In 1992, 15 populations (Villanueva de Jara, Monte Oscuro, Pancrudo, *coerulea*, Flamande, Provence, Marais de Luçon, Europe, Maktar, *romanica*, *quasifalcata*, Anik, Maron, Krasnokutskaya, *glomerata*) were planted in the nursery (1 block, 30–50 plants per population, 70 cm between plants) at Station d'amélioration des plantes fourragères INRA (Lusignan, France). In 1993, the following traits were recorded on each plant: rhizomatous plants (plants with lateral expansion of the crown by underground, horizontal stems, as described by Heinrichs in 1963); diameter of the plants at their base in cm (on 19/03 and 07/08, mean of 2 perpendicular measurements per plant); 8 consecutive measurements of plant height from 19/03 to 27/04/1993; 8 consecutive measurements of the length of 1 tagged stem per plant from 16/04 to 16/06/1993. Two cuttings were made (on 28/06 and 10/08/1993) and an average fresh weight per plant was estimated for each cutting.

In 1993, 21 populations (among the 25 populations, only the 4 Spanish landraces were omitted) were planted at INRA (Lusignan, France) in the nursery (3 blocks, 19 plants per block, 70 cm between plants). In September 1993, the individual measurements on each plant were: growth habit (from 1 (prostate) to 5 (erect), stem size (from 1 (thin) to 3 (thick), leaf area (from 1 (small) to 3 (large), inflorescence shape (1 (globular), 2 (moderately elongated), and 3 (elongated). Three scores for regrowth were recorded per

Table I. Origin of the 25 populations.

Name	*Subspecies*	*Country*	*Region*	*Type of population*	*Type or use*	*Ploidy*	*Longitude*	*Latitude*	*Altitude (m)*
Flamande	*sativa*	France	North	Landrace	Flemish	4 x			
Europe	*sativa*	France	–	Variety	Flemish	4 x			
Provence	*sativa*	France	Provence	Landrace	Provence	4 x			
Marais de Luçon	*sativa*	France	Vendée	Landrace	Flemish	4 x			
Magali	*sativa*	France	–	Variety	Provence	4 x			
D15	*sativa*	Morocco	Draa valley	Landrace	Pre-Saharian oasis	4 x	30.48N	5.35W	–
Dem3	*sativa*	Morocco	Demnate	Landrace	Valley	4 x	31.45N	7.06W	–
Pool 5	*sativa*	Morocco	Atlas	Pool	Mountain	4 x			
Gabès	*sativa*	Tunisia	–	Landrace	Irrigated oasis	4 x	32.57N	10.02E	–
Maktar	*sativa*	Tunisia	Maktar	Landrace	–	4 x	–	–	450
Aragon	*sativa*	Spain	Cuenca	Landrace	–	4 x	41.40N	0.50W	250
Mediterraneo	*sativa*	Spain	Murcia	Landrace	–	4 x	37.55N	1.15W	100
Tierra de Campos	*sativa*	Spain	Palencia	Landrace	–	4 x	41.35N	4.40W	700
Ampurdan	*sativa*	Spain	Gerona	Landrace	–	4 x	42.10N	3.10E	70
Villanueva de Jara	*sativa*	Spain	Cuenca	Wild	Rangeland	4 x	39.26N	1.58W	730
Monte Oscuro	*sativa*	Spain	Zaragoza	Wild	–	4 x	40.45N	1.16E	400
Pancrudo	*sativa*	Spain	Teruel	Wild	–	4 x	40.46N	1.02W	1 100
coerulea	*sativa*	Russia	Daghestan	Wild	Arid steppes	2 x	42.00N	47.00E	–
Maron	*falcata*	France	Lorraine	Wild	–	4 x	54.00N	4.20E	215
Malzeville	*falcata*	France	Lorraine	Wild	–	4 x	48.41N	6.31E	212
Krasnokutskaya	*falcata*	Ukraine		Wild	–	4 x			
Anik	*falcata*	Canada	Alberta	Variety	–	2 x			
romanica	*falcata*	Russia	Krasnodar?	Wild	Arid steppes	2 x	45.02N	39.00E	–
quasifalcata	*falcata*	Russia	Krasnodar?	Wild	Meadows	2 x	45.02N	39.00E	–
glomerata	*glomerata*	France	Vaucluse	Wild	–	2 x	43.30N	5.40E	450

block (from 1 (weak regrowth) to 5 (strong regrowth), on 28/07, 08/10 and 26/10/1993). In 1994, the following traits were recorded per plant: diameter of the plants (mean of 2 perpendicular measurements, 15/03/1994), 5 measurements of plant height from the 08/04 to 10/05/1994, flower colour (1 (pure purple), 2 (variegated), 3 (pure yellow). The percentage of rhizomatous plants per population, and the percentage of survival after winter were recorded. Flowering data was noted according 2 criteria: 10 and 50% flowered plants per block. The third inflorescence of 10 plants per block was sampled in order to evaluate pod setting (percentage of flowers that set pods), number of seeds per pod and mean seed weight.

In 1993, the 25 populations were sown in small 2-row plots 2.60 m long, 0.30 m apart, with 4 blocks. Two cutting managements were used in 1994, each on 2 blocks, one with frequent cuttings (dates of cuttings: 10/05, 23/06, 04/08, 16/09, 08/11), the other with normal cuttings (dates of cuttings: 24/05, 02/07, 19/08, 04/10). For each cutting, in each block and for each genotype, dry matter yield was evaluated. For the frequent and normal cutting regimes, 19 and 15 stand height measurements respectively were made between 13/03/1994 and 28/11/1994 during the successive regrowths.

Analyses of variance were performed for all recorded traits. The data were analysed by trial because all the populations were not represented in each trial and this reduced the possibilities of multivariate analysis. For the 1992 nursery, the effect of population and the effect of plants nested within population were tested. For the 1993 nursery, the block effect was added to the model. For the trial in small plots, the effects of population and block were tested, and the data recorded under the 2-cutting managements were considered as separate traits. Correlations between characters were computed on the mean values of the populations, and principal component analyses (PCA) were performed. The objective was to investigate the relationships between populations and to structure the variability in groups. Euclidian distances between populations were computed from their coordinates on the principal components, and an ascendant hierarchical clustering was carried out using the complete linkage method. The partition observed from the view of the tree was represented on the plan of the first 2 components.

RESULTS

Nursery 1992

For all characters, the population effect was highly significant ($P < 0.001$). The effect of plants nested within population, tested against population effect, was not significant. PCA was computed with all the available traits. The first component accounted for 83.4% of the variation, the second component 11.3%. The first component was positively correlated with plant height, stem length and yield. The second component was defined by diameter and percentage of rhizomatous plants. The tree based on Euclidian distances on principal components showed that 2 groups of populations can be defined; one group consisted of landraces or varieties of the ssp *sativa* (Maktar, Europe, Provence, Flamande and Marais de Luçon) and the tetraploid *falcata* population Krasnokutskaya, and the other group was the remaining populations, most of which were wild (fig 1). *Sativa* landraces and varieties were characterised by tall plants, long stems, high yields and no rhizomatous plants. The other populations, diploid or tetraploid *falcata* populations, the diploid *sativa* population, the diploid *glomerata* populations and tetraploid wild *sativa* populations *Mielga*, were characterised by lower plant height, stem length and yields (especially for the second cutting in August) and by the presence of rhizomatous plants (table II). However, in this last group, the populations showed large variation for plant diameter and percentage of rhizomatous plants. Diploid wild populations (*coerulea*, *romanica* and *quasifalcata*) had a low percentage of rhizomatous plants and a small plant diameter.

Nursery 1993

For all traits, the population effect was significant ($P < 0.01$), and the effect of block was either non-significant or lower than population effect.

With the characters available for all populations in 1993 and 1994 (flowering dates, plant diameter and rhizomatous trait were discarded as missing for either Gabès or *glomerata*). PCA

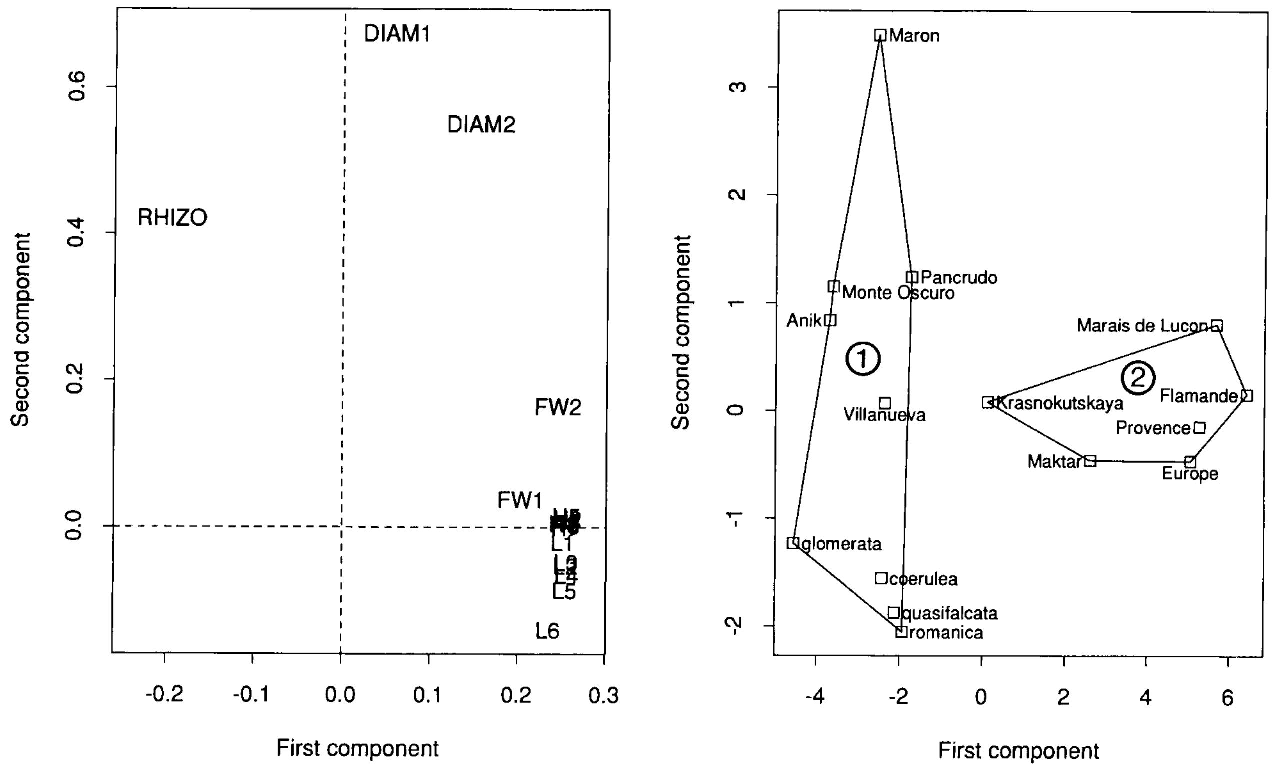

Fig 1. PCA diagram on the 1–2 plane of 15 populations studied in 1993 in the 1992 nursery. Left: diagram of the coefficients of correlation of the characters with the first 2 components; Right: diagram of the 15 populations. The characters considered were plant heights (H1–H6), stem lengths (L1–L6), plant diameters (DIAM1, DIAM2), % rhizomatous plants (RHIZO), fresh weight per plant in 2 cuttings (FW1, FW2). **1** and **2** are the 2 groups shown by partition from Euclidian distances on the components of PCA.

Table II. Main characteristics of the populations planted in 1992 in the nursery and studied in 1993 (SE: standard error).

Population	*Type* [a]	*Diameter (cm) 07/08/93*	*Plant height (cm) 01/04/93*	*Stem length (cm) 07/05/93*	*% rhizomatous plants*	*Plant fresh cutting 28/06/93*	*Yield cutting 10/08/93*
Flamande	C 4 x	44.2	27.3	64.2	0.00	1.672	0.791
Europe	C 4 x	38.6	23.8	60.7	0.00	1.419	0.659
Provence	C 4 x	40.2	24.5	60.6	0.00	1.386	0.824
Maktar	C 4 x	35.9	17.3	50.1	0.00	1.003	0.648
Marais de Luçon	C 4 x	47.7	25.9	59.2	0.00	1.950	0.771
Villanueva	W 4 x	32.7	4.1	31.5	51.22	0.974	0.243
Monte Oscuro	W 4 x	37.6	2.0	29.4	74.29	0.713	0.083
Pancrudo	W 4 x	40.6	5.6	31.8	56.76	1.106	0.345
coerulea	W 2 x	23.6	6.5	31.2	23.40	0.936	0.053
Maron	W 4 x	46.8	4.7	27.3	91.30	1.153	0.382
Krasnokutskaya	W 4 x	35.3	10.7	39.9	35.71	1.313	0.453
Anik	C 2 x	31.8	3.1	25.6	77.50	0.821	0.173
romanica	W 2 x	23.7	4.3	36.9	10.20	1.273	0.147
quasifalcata	W 2 x	24.3	4.8	33.1	4.08	1.383	0.149
glomerata	W 2 x	21.7	0	25.5	62.50	0.363	0.031
Mean		34.33	10.97	40.5	32.46	1.164	0.384
SE		12.05	5.12	7.25	41.96	–	

[a] C: cultivated; W: wild; 4 x: tetraploid; 2 x: diploid.

gave 2 components accounting for 72.5 and 11.7% of variation respectively. The first component was explained by regrowth scores, growth habit, stem size, leaf size, inflorescence shape, plant height, number of seeds per pod and mean seed weight. The second component was explained by winter survival (fig 2). The Euclidian distances isolated the group of *sativa* landraces and varieties from the remaining populations by their higher regrowth scores and their higher plants. The Ukrainian population Krasnokutskaya was intermediate between these 2 groups, and *glomerata* had lower regrowth scores and plant heights than the other wild populations (table III). Some landraces from the south of the Mediterranean (Gabès, D15, Pool5) had a lower winter survival than French landraces and populations (Flamande, Europe, Marais de Luçon, Provence and Magali) (table III). In this diagram, among the wild populations, the *sativa* Mielga cannot be distinguished from the others. The distinction between Mielga and other wild populations was obvious considering flower colour (table III); Mielga populations and *coerulea,* as the other *sativa* populations, had purple flowers, whereas *falcata* populations had yellow flowers. Pod set showed a wide variation; wild diploid (*coerulea*, *romanica* and *quasifalcata*) and Mielga (Villanueva, Monte Oscuro and Pancrudo) populations had lower pod sets than the remaining ones (table III). Within the wild populations, the diploid populations had smaller seeds than the tetraploid populations (on average, mean seed weight was 1.34 (± 0.32) mg for diploid populations, and 1.71 (± 0.27) mg for tetraploid wild populations). French landraces and varieties flowered earlier than the other populations (table III). Flowering dates of *coerulea*, *romanica* and *quasifalcata* were late.

Small plots 1993

The population effect was significant for the stand heights and yields per cutting.

A PCA was computed on the measurements of stand heights (19 and 17 measurements in frequent and normal cutting regimes, respectively), and on the yield evaluation per cutting and on the total (5 cuttings in the frequent regime; 4 cuttings in the normal regime). The first component explained 81.1% of the variation, the second component 14.2%. All height measurements and

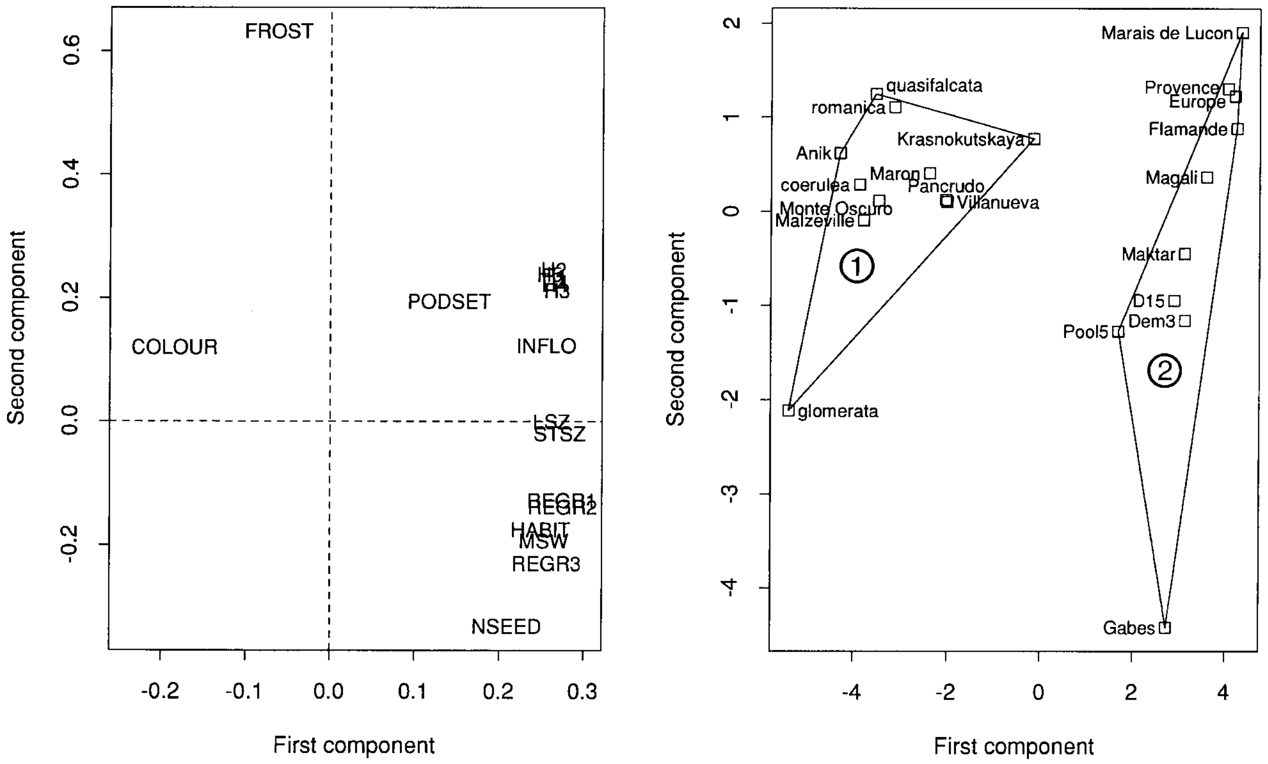

Fig 2. PCA diagram on the 1–2 plane of 21 populations studied in 1993 and 1994 in the 1993 nursery. Left: diagram of the coefficients of correlation of the characters with the first 2 components; Right: diagram of the 21 populations. The characters considered were growth habit (HABIT), regrowth scores (REGR1 to REGR3), stem size (STSZ), leaf size (LSZ), inflorescence shape (INFLO), plant heights (H1–H5), number of seeds per pod (NSEED), mean 1 000 seed weight (MSW), frost damage (FROST), pod set (PODSET), flower colour (COLOUR). **1** and **2** are the 2 groups shown by partition from Euclidian distances on the components of PCA.

yields per cutting except on the first cutting contributed to the first component. The second component was correlated positively to yield of the first cutting in both regimes, and negatively to yield of autumn cutting. Two main groups appeared with the Euclidian distances (fig 3), one with the cultivated *sativa* populations or varieties that were high yielding and tall, the other with the wild populations (*falcata*, *sativa* Mielga and *coerulea*, and *glomerata*) that were lower yielding and shorter (table IV). Within the cultivated populations, the French populations adapted to a cool climate were separated from the Mediterranean populations because of their higher yield in the first cutting and lower relative yield in autumn. The *glomerata* population was close to the group of wild populations, but with the lowest yield and weak growth, and a low yield in first cutting. In autumn, forage yield was low for all the wild populations (table IV). For the cultivated populations, yield and stand height in autumn were similar for north European populations and for Mediterranean populations, but the proportion of yield produced in autumn over the annual yield was higher for the Mediterranean populations than for the others.

DISCUSSION

Whatever the morphological and agronomic traits used to describe the populations, 2 main groups of populations were separated. The 3 trials gave consistent information on the populations studied. The first group contained the French, Spanish and North African varieties and populations. They were characterised by high yields, tall plants, erect growth habit, good regrowth after cutting, and high mean seed weight. The French populations (Europe, Flamande and Marais de Luçon) yielded more in the moderately cool climate of Lusignan than the Mediterranean climates (Provence, Magali, Dem3, Pool5, D15, Gabès, Tierra de Campos, Ampurdan, Mediterraneo and Aragon), and had purple flowers variegated with yellow instead of pure purple. A large proportion of annual forage yield was pro-

Table III. Main characteristics of the populations planted in 1993 in the nursery and studied in 1993 and 1994.

Population	*Type*	*Regrowth score* [a]	*Growth habit* [b]	*Winter survival* [c]	*Plant height (cm)* [d]	*Flowering date* [e]	*Flower colour* [f]	*Mean seed weight (mg)*	*Pod set*
Flamande	C 4 x	3.83	3.00	96.3	46.8	2	1.15	2.33	48.33
Europe	C 4 x	3.33	2.93	100	47.7	3	1.05	2.36	60.43
Provence	C 4 x	3.67	3.00	100	46.3	7	1.16	2.24	55.97
Marais de Luçon	C 4 x	3.33	3.00	100	53.8	7	1.19	2.23	52.03
Magali	C 4 x	3.50	3.00	100	43.7	6	1.07	2.54	46.67
D15	C 4 x	3.67	3.00	83.9	37.9	13	1.13	2.19	36.90
Dem3	C 4 x	4.17	3.00	91.2	31.8	13	1.06	2.59	51.30
Pool 5	C 4 x	3.17	3.00	78.6	30.4	11	1.02	2.29	48.60
Gabès	C 4 x	3.83	3.00	32.1	26.7	–	1.00	2.53	47.50
Maktar	C 4 x	4.00	3.00	92.7	38.7	8	1.05	2.38	44.10
Villanueva	W 4 x	1.83	2.07	100	22.7	10	1.16	1.73	37.67
Monte Oscuro	W 4 x	1.17	1.32	100	21.0	13	1.00	1.53	26.80
Pancrudo	W 4 x	1.67	1.91	100	23.7	5	1.00	1.60	29.47
coerulea	W 2 x	1.33	1.32	100	16.5	25	1.00	0.89	43.73
Maron	W 4 x	1.00	1.21	98.2	18.0	8	2.91	1.63	56.00
Malzeville	W 4 x	1.50	1.21	100	12.0	11	2.93	1.56	53.77
Krasnokutskaya	W 4 x	1.50	2.51	98.2	29.7	10	2.92	2.20	53.00
Anik	C 2 x	2.33	1.24	100	11.2	15	3.00	1.43	52.47
romanica	W 2 x	1.33	1.78	100	20.5	19	3.00	1.34	40.57
quasifalcata	W 2 x	1.17	1.38	100	18.8	22	3.00	1.34	36.87
glomerata	W 2 x	1.00	2.52	81.8	5.3	–	3.00	1.72	21.10
Mean		2.49	2.31	92.9	28.7	11	1.70	1.90	44.86
SE		0.39	0.17	15.4	3.21	3.2	0.08	0.19	9.65

[a] Score from 1 (no regrowth) to 5 (strong regrowth) (06/10/1993); [b] score from 1 (prostrate) to 3 (erect) (02/09/1993); [c] % of surviving plants after winter (07/03/1994); [d] measured on 02/05/1994; [e] measured when 10% of the plants had one open flower, in days after 18/05/1994; [f] score: 1 (purple), 2 (variegated), and 3 (yellow).

duced in autumn in the Mediterranean populations, and this feature confirms the regrowth capacity of Mediterranean types in autumn compared to northern European types and wild populations.

The second group contained wild *falcata* (Maron, Malzeville, *romanica*, *quasifalcata* and Krasnokustkaya) and wild *sativa* (*coerulea*, Monte Oscuro, Villanueva and Pancrudo) populations including the *falcata* Canadian variety Anik, and were characterised by reduced growth, low forage yields, prostrate growth habit, low mean seed weight. Their yield in autumn was low or zero. The *glomerata* population was close to this group. Kranokutskaya tended to be intermediate between the main 2 groups because it had better regrowth scores and higher yields than the other *falcata* populations. The Mielga populations, known as wild *sativa*, could not be distinguished morphologically and agronomically from the *falcata* populations, except by flower colour, purple for Mielga, yellow for *falcata*. The wild Mielga populations were clearly different from cultivated populations, as that were already shown by Delgado Enguita (1989) and Prosperi *et al* (1993). They could represent the wild populations that were already present in Spain when the cultivated forms were introduced by the Arabs in the 8th Century (Delgado Enguita, 1989). The differences between wild Spanish populations and cultivated ones show the progress made by man in domesticating lucerne. The main improvements related to a more erect growth habit, adapted to forage harvesting, regrowth and forage yield, but also pod set and mean seed weight. It seems that very few crosses occurred between Mielga and cultivated landraces, even though they are sympatric (Prosperi *et al,* 1990). This might be the consequence of the crop management of the landraces, often irrigated, and harvested before flowering. A large range of flowering dates was observed. Response to temperature and photo-

Table IV. Total yield in 1994 (t/ha), yield and stand height (cm) in the first cutting in spring (10/05), yield and stand height in the last cutting in autumn (08/11) in frequent cutting regime of the small plot trial sown in 1993.

Population	*Type*	*Total yield (t/ha)*	*Yield of spring cutting (t/ha)*	*Height at spring cutting (cm)*	*Yield of autumn cutting (t/ha)*	*Height at autumn cutting (cm)*
Flamande	C 4 x	21.04	6.57	93.75	1.81	45.50
Europe	C 4 x	19.94	7.57	92.15	1.50	44.00
Provence	C 4 x	16.11	4.89	86.25	1.62	46.75
Marais de Luçon	C 4 x	17.11	6.23	88.75	1.25	37.50
Magali	C 4 x	18.44	5.45	81.25	1.86	43.75
D15	C 4 x	16.38	4.61	75.00	1.71	51.75
Dem3	C 4 x	10.80	2.24	60.00	1.66	49.75
Pool 5	C 4 x	10.55	2.80	56.25	1.34	49.25
Gabès	C 4 x	5.56	1.08	52.50	0.81	52.00
Maktar	C 4 x	15.60	4.41	78.75	1.58	48.25
Aragon	C 4 x	11.96	2.77	77.50	1.55	49.25
Mediterraneo	C 4 x	10.26	2.85	68.75	1.13	46.50
Tierra de Campos	C 4 x	13.98	3.68	80.00	1.30	43.75
Ampurdan	C 4 x	10.90	3.09	65.00	1.56	40.50
Villanueva	W 4 x	7.67	2.98	58.75	0.00	7.25
Monte Oscuro	W 4 x	4.97	2.65	50.00	0.00	1.75
Pancrudo	W 4 x	6.50	3.13	55.00	0.00	2.50
coerulea	W 2 x	5.84	3.50	57.50	0.00	1.50
Maron	W 4 x	11.08	5.15	63.75	0.05	6.25
Malzeville	W 4 x	9.52	3.94	62.50	0.06	8.00
Krasnokutskaya	W 4 x	12.28	6.08	67.50	0.00	8.25
Anik	C 2 x	5.12	2.64	43.75	0.00	1.00
romanica	W 2 x	6.84	4.03	58.75	0.00	4.25
quasifalcata	W 2 x	6.52	4.15	53.75	0.00	4.50
glomerata	W 2 x	0.30	0.28	31.25	0.00	1.75
Mean		11.01	3.87	66.30	0.83	27.82
SE		1.79	0.77	6.93	0.19	3.35

period of each genotype could be involved in flowering (Guy *et al,* 1971).

The variegated flower colour of the French populations provides evidence for the genetic introgression of *falcata* types in these *sativa* populations. The crosses between *sativa* and *falcata* should have occurred in northern and eastern France and Europe where both sub-species coexist (Lesins and Lesins, 1979). The *falcata* trait useful for adaptation of lucerne to the northern climates of Europe was their winter resistance associated with a reduced growth during cold periods.

The convergent evolution of forms between wild *falcata* and wild *sativa* Mielga probably shows the strong advantages of the prostrate growth habit, weak and slow growth for adaptation and persistence in nature. Thanks to this morphology, the vegetative buds escape animal grazing and the plants resist animal treading. The prostrate growth habit and rhizomatous trait were shown to contribute to tolerance to grazing (Prosperi *et al,* 1993), so that the wild tetraploid populations with these traits could be used in crosses with elite cultivars to improve their persistence under grazing.

Within the wild types, no distinction was obvious between diploid and tetraploid populations. The *romanica* and *quasifalcata* populations (2 x) are not different from Maron, Malzeville and Krasnokutskaya (4 x), and *coerulea* (2 x) are not different from Mielga (4 x). Only mean seed weight was higher on wild tetraploids than on wild diploids. The theoretical advantage of tetraploid forms on diploid ones for growth, yield and development was inapparent. Furthermore, ploidy level is considered as a barrier against interbreeding, thus allowing divergent evolution (Lesins and Lesins, 1979). This genetic isolation between diploid and tetraploid had no conse-

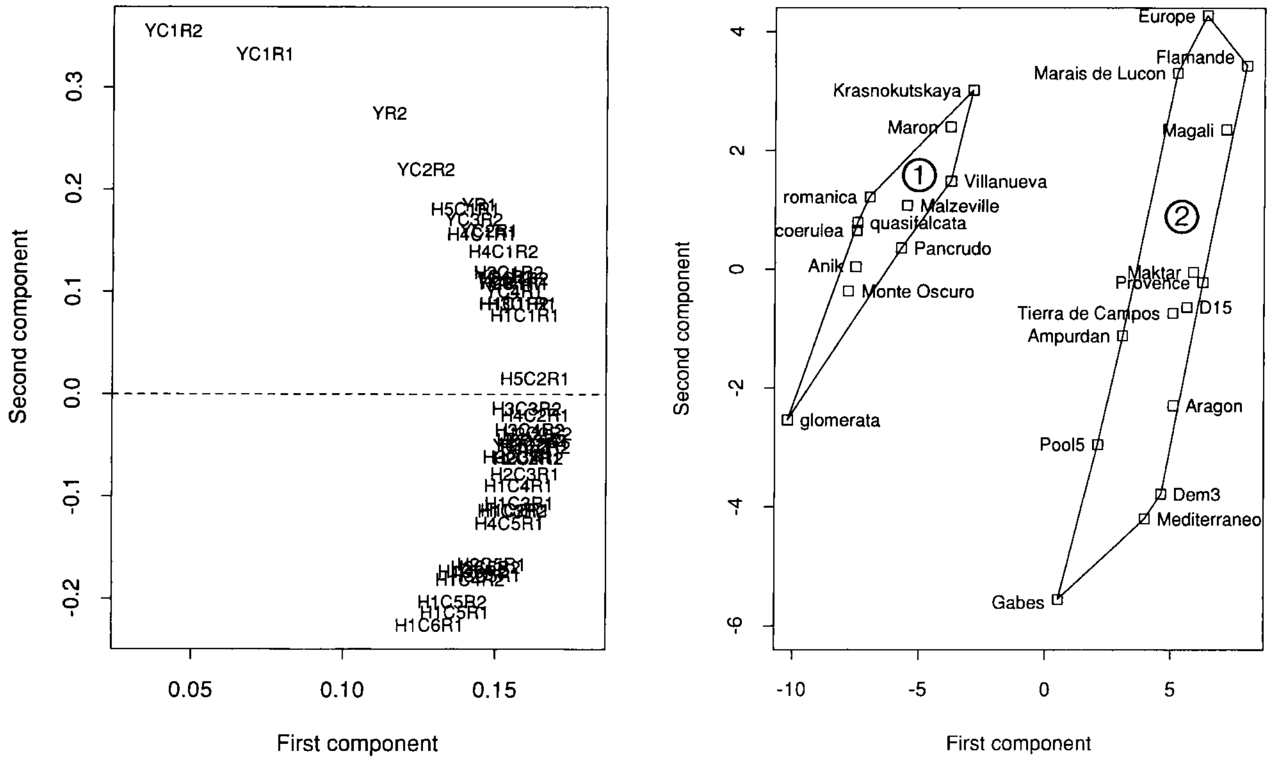

Fig 3. PCA diagram on the 1–2 plane of 25 populations studied in 1994 in the 1993 small plot trial. Left: diagram of the coefficients of correlation of the characters with the first 2 components; Right: diagram of the 25 populations. The characters considered were the measurements of plants heights (H1–H6) per regrowth cycle (C1–C6) in the 2 cutting regimes (R1, R2) and yields per cutting (Y) in the 2 cutting regimes. **1** and **2** are the 2 groups shown by partition from Euclidian distances on the components of PCA.

quence on the morphological traits studied here. However, the fact that cultivated lucernes are tetraploid proves the higher potentialities of certain tetraploid populations.

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REFERENCES

Birouk A, Guy P (1986) Partition de populations marocaines de luzerne locale en vue de leur conservation en réservoir massal. *In: Distance, divergence et variabilité génétique,* Séminaire du département de génétique et d'amélioration des plantes, Méribel *11-15 mars 1986,* INRA, Paris, France, 196-203

Birouk A, Dattee Y, Sadiki M, Roumet P (1989) Évaluation agronomique et adaptation de populations marocaines de luzerne (*Medicago sativa* L). *agronomie* 9, 363-376

Bolton JL, Goplen BP, Baenziger H (1972) World distribution and historical developments. *In: Alfalfa Science and Technology* (CH Hanson, ed), ASA Publishers, *Agronomy (Madison)* 15, 1-34

Delgado Enguita I (1989) Estudio de la variabilidad de las Mielgas aragonesas en areas de precipitacion anual inferior a 600 mm. Thesis doctoral, Escuela Technica Superior de Ingeniero Agronomos, Universidad Politecnica de Madrid, Spain, 168 p

Dunbier MW, Eskew DL, Bingham ET, Schrader LE (1975) Performance of genetically comparable diploid and tetraploid alfalfa: agronomic and physiological parameters. *Crop Sci* 15, 211-214

Guy P, Blondon F, Durand J (1971) Action de la température et de la durée d'éclairement sur la croissance et la floraison de 2 types éloignés de luzerne cultivée, *Medicago sativa* L. *Ann Amélior Plant* 21, 409-422

Heinrichs DH (1963) Creeping alfalfas. *Adv Agron* 15, 317-337

Hidalgo Maynar F (1966) Clasificatión de las alfalfas españolas. Association de Investigation para la mejora de la alfalfa, Zaragoza, Spain, 86 p

Lesins KA, Lesins I (1979) *Genus* Medicago *(Leguminosae) A taxonomic study.* Junk bv Publishers, The Hague, The Netherlands, 228 p

Prosperi JM, Angevain M, Mansat P (1990) Valorization of forage genetic resources for selection of adapted cultivars. Example of wild

Mediterranean lucernes. *6th Meeting FAO on Mediterranean Pastures and Fodder Crops, Bari,* 95-98

Prosperi JM, Angevain M, Génier G, Olivieri I, Mansat P (1993) Sélection de nouvelles légumineuses fourragères pour les zones difficiles méditerranéennes. *Fourrages* 135, 343-354

Quiros CF, Bauchan GR (1988) The genus *Medicago* and the origin of the *Medicago sativa* complex. *In: Alfalfa and Alfalfa Improvement* (AA Hanson, ed), ASA-CSSA-SSSA Publishers, *Agronomy (Madison)* 29, 93-124

Volenec JJ (1988) Herbage growth and carbohydrate metabolism of diploid and tetraploid alfalfa. *Crop Sci* 28, 128-132