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Influence of age in germination of seeds of *Lotus* (*Fabaceae*)

Abstract

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Germination of seeds from 179 populations belonging to 10 taxa of *Lotus* L. sect. *Lotus* collected in Spain, Portugal, Italy and France is studied. Seeds of different ages were sown to check where germination percentage and germination dynamics could change with age. Results of this paper indicate that time elapsed by seeds to start germination is rather short. Values of t_{50} (time elapsed to complete 50 % germination of the total germinated seeds) shown a general decrease with age both in annual and perennial taxa, more pronounced in the former than in the later. Results demonstrate that there are clear differences in germination percentages and germination dynamics between annual and perennial taxa. Some hypotheses are given to explain these differences.

Introduction

Lotus includes c. 190 species from temperate areas all around the world, with a main diversification centre in the Mediterranean Region (Polhill 1981).

The fruit is a dehiscing legume which produces two-four seeds in annuals (*L. parviflorus*, *L. castellanus* or *L. hispidus*, by instance) and more than ten in perennials. Out of the W Mediterranean the species with the highest seed production is *L. pedunculatus* with 18-20 seeds per legume and between 2000 and 3000 seeds by a well grown specimen.

Differences in germination capacity amongst populations within a particular taxon and variations according to age are fairly well known (Anderson & Milberg 1998), and seed germination capacity has been used as a taxonomic tool to reinforce taxa separation, as in *Teucrium* (Gramma-Ghrabi & al. 1989) or *Asphodelus* (Díaz Lifante 1994).

No less than 15 families of Angiospermae, including *Fabaceae*, have physical seed dormancy (Mujica & Rumi 1993; Baskin & Baskin 1998), which in *Fabaceae* is owed to seed-coat structure, impermeable to water, which results in low germination of seeds, which are classified as hard.

There are many studies dealing with seed germination in Lotus, but non of them have been addressed to taxonomy. Hurt & Nelson (1985) indicated that in *L. corniculatus* the highest germination percentages were reached by untreated seeds at 20 ºC. Eight years later Mujica & Rumi (1993) studied seed germination on *L. tenuis* at different temperatures and demonstrated that the highest germination percentages and more rapid germination were obtained at 21 ºC.

This study includes observations on germination behaviour of different W Mediterranean taxa of *Lotus* L. sect. *Lotus*. Total germination percentage, germination dynamics in relation to life cycle and seed viability and dormancy at different ages are included.

Material and methods

Ten taxa belonging to five species of *Lotus* sect. *Lotus* from Iberian Peninsula, French Alps and Apeninos Mountains (Italy) are studied. Two taxa, *L. hispidus* and *L. castellanus*, are annuals, and the remaining perennials. Seeds from 179 populations were collected between 2000 and 2002 and a total of 16438 seeds were sown.

The number of populations and seeds for each taxon, as well as their life cycle are indicated in table 1. The origin of each population is indicated in the appendix. Only *L. corniculatus* subsp. *glacialis*) is endemic of the Iberian Peninsula (Valdés 2000).

Mature seeds from each population were collected and stored in paper bags at room temperature. Seeds were always sown in December.

Table 1. Taxa, number of populations (N) and number of seeds (n) studied. L.c., life cycle.

Taxa	N	n	L.c.
L. corniculatus L. subsp. *corniculatus*	29	2661	perennial
L. corniculatus subsp. *alpinus* (DC.) Rothm.	3	160	perennial
L. corniculatus subsp. *carpetanus* (Lacaita) Rivas Mart.	16	1604	perennial
L. corniculatus subsp. *glacialis* (Boiss.) Valdés	7	591	perennial
L. corniculatus subsp. *delortii* (Timb.-Lagr.) O. Bolòs & Vigo	3	335	perennial
L. corniculatus subsp. *preslii* (Ten.) P. Fourn.	6	615	perennial
L. glaber Mill.	32	2205	perennial
L. pedunculatus Cav.	61	6050	perennial
L. castellanus Boiss. & Reut.	12	1213	annual
L. hispidus Desf. ex DC.	10	1004	annual

To study germination capacity in relation to age, as well as possible differences in viability and germination dynamics of seeds, these were sown at three different ages: at the collecting year (age 0), one year old (age 1) and two year old seeds (age 2).

Whenever possible, 100 seeds from each population in four replicates of 25 were sown in Petri dishes with two layers of filter paper soaked with distillate water. Petri dishes were wrapped with paraphine film and incubated to germinate.

As the highest germination percentages were obtained in *L. corniculatus* and *L. tenuis* at 20 ºC with constant temperature, as indicated above (Hurt & Nelson 1985; Mujica & Rumi 1993), experiments have been carried out at this constant temperature. Photoperiod has always been of eight hours light and 16 hours darkness. This regime of light-darkness allowed seedlings to survive, which can later be used for karyological, reproductive and genetic studies.

A seed was considered as germinated when the radicle was visible. Germinated seeds were counted daily during the first month, every other day the following two weeks and once a week during the rest of the experiment. To analyse germination dynamics two parameters have been considered: t_0 or time between sowing and first germination and t_{50} as time necessary to reach 50 % of total germination percentage. For each sample mean, standard deviation and variation interval were evaluated.

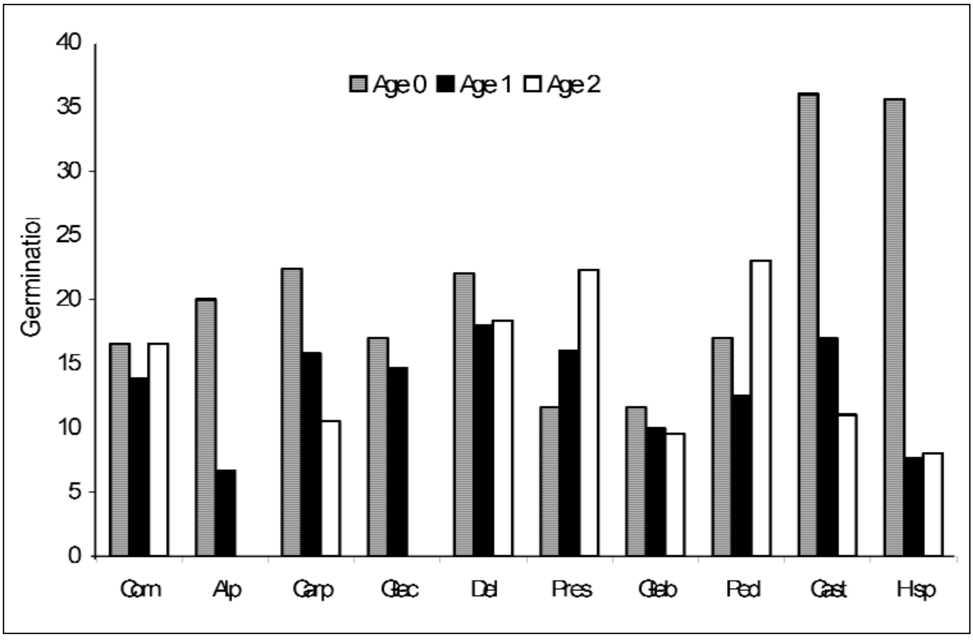

Fig. 1. Germination percentages in three consecutive years. **Cast,** *L. castellanus*; **Hisp,** *L. hispidus*; **Glac,** *L. corniculatus* subps. *glacialis*; **Del,** *L. corniculatus* subsp. *delortii*; **Alp,** *L. corniculatus* subsp. *alpinus*; **Carp,** *L. corniculatus* subsp. *carpetanus*; **Ped,** *L. pedunculatus*; **Corn,** *L. corniculatus* subsp. *corniculatus*; **Glab,** *L. glaber*; **Pres,** *L. corniculatus* subsp. *preslii*.

Table 2. Total germination percentages. **N,** number of populations; $\overline{X}$ **,** mean; **σ,** standard deviation; **v.i.,** variation interval.

Taxon	**Age 0** N	$\overline{X}$	σ	v.i.	**Age 1** N	$\overline{X}$	σ	v.i.	**Age 2** N	$\overline{X}$	σ	v.i.
L. corniculatus subsp. *corniculatus*	12	16,52	10,14	4,00 - 31,25	20	13,78	7,93	3,00 - 38,00	2	16,50	4,95	13,00 - 20,00
L. corniculatus subsp. *alpinus*	1	20,00			2	6,67	3,77	4,00 - 9,33				
L. corniculatus subsp. *carpetanus*	7	22,35	17,18	3,33 - 46,00	10	15,77	11,90	2,00 - 36,00	2	10,50	7,78	5,00 - 16,00
L. corniculatus subsp. *glacialis*	7	16,96	6,21	10,00 - 26,00	2	14,67	3,77	12,00 - 17,33				
L. corniculatus subsp. *delortii*	2	22,00	14,14	12,00 - 32,00	1	18,00			1	18,33		
L. corniculatus subsp. *preslii*	4	11,58	14,51	4,00 - 33,33	2	16,00	9,90	9,00 - 23,00	2	22,27	5,10	25,88 - 18,66
L. glaber	4	11,58	14,51	4,00 - 33,33	20	9,95	7,38	2,00 - 33,00	2	9,50	4,95	6,00 - 13,00
L. pedunculatus	26	16,96	11,54	4,00-51,00	43	12,44	10,29	3,00 - 54,00	2	23,00	9,90	16,00 - 30,00
L. castellanus	2	36,00	5,66	32,00 - 40,00	11	16,95	9,48	3,00 - 38,46	2	11,00	11,31	3,00 - 19,00
L. hispidus	3	35,56	24,64	22,69 - 64,00	7	7,57	4,47	2,00 - 14,00	2	8,00	5,66	4,00 - 12,00

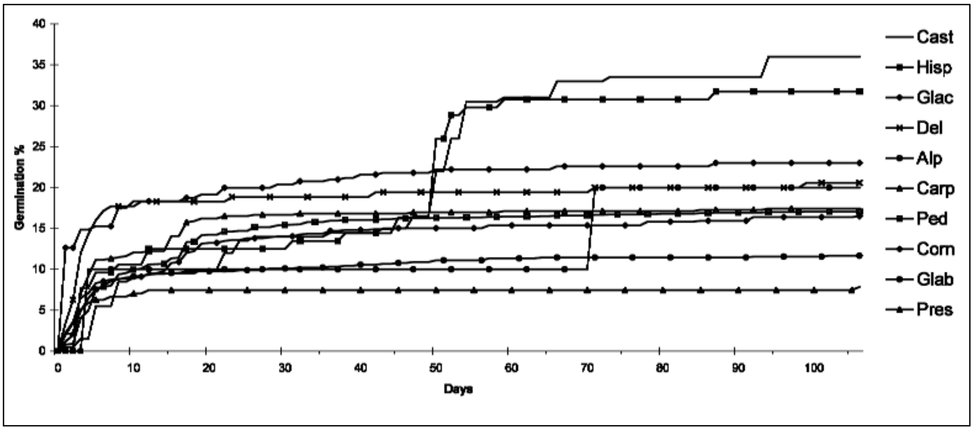

Fig. 2. Germination dynamics in seed of age 0. For abbreviations, see fig. 1.

Two different statistical analysis were done based on seeds of age 0 to get information independent from age. In the first analysis total germination, t_0 and t_{50} were analysed taken into consideration procedence of populations. In the second, the same parameters are used but searching for potential correlations amongst taxa. As only samples with more than two replicates were considered, *L. corniculatus* subsp. *preslii*, *L. corniculatus* subsp. *alpinus* and *L. hispidus* were excluded. Kruskal-Wallis proof (p <.05) for no parametric data was developed by using SPSS ver. 11.5 statistical programme.

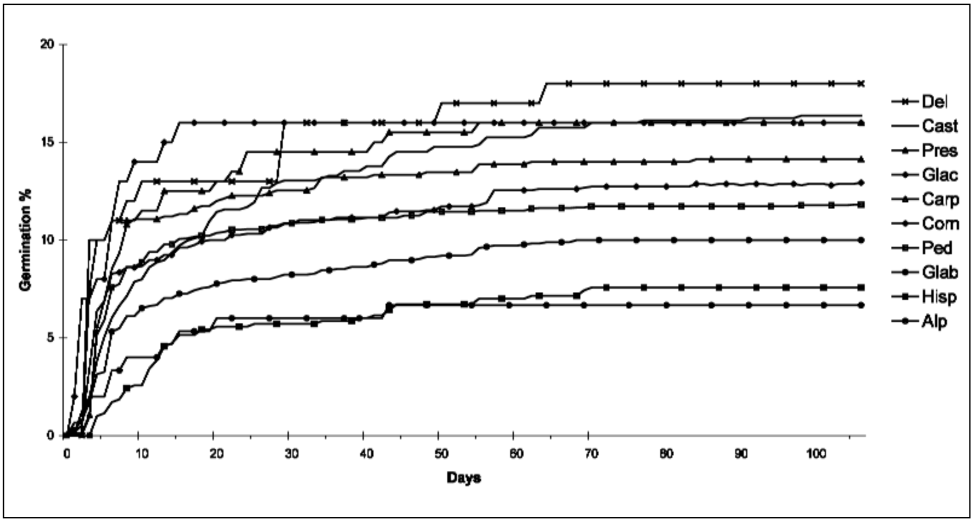

Fig. 3. Germination dynamics in seeds of age 1. For abbreviations, see fig. 1.

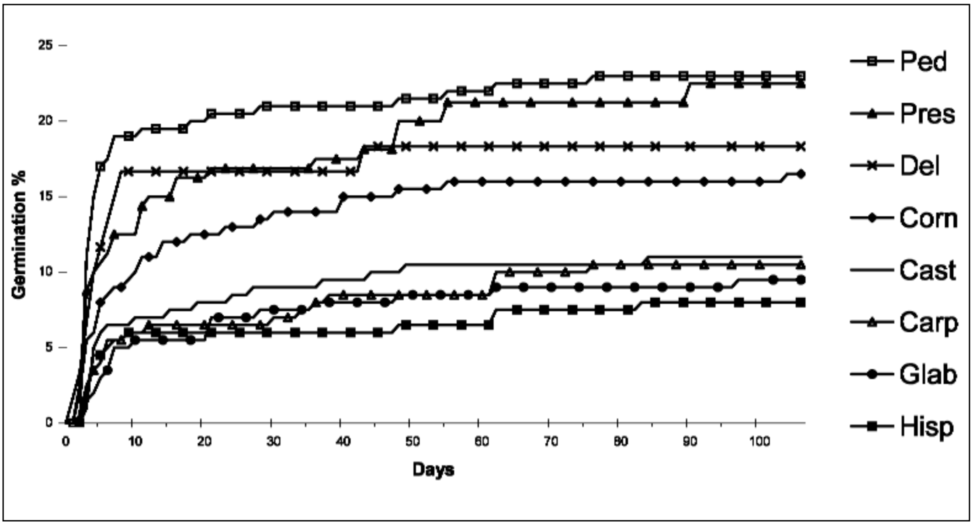

Fig. 4. Germination dynamics in seeds of age 4. For abbreviations, see fig. 1.

Results

Numbers of populations and seeds used, mean of germination percentages, standard deviation and variation interval for each taxa at different ages are shown in table 2. Figure 1 graphically shows these percentages.

Germination percentages are always lower than 50 % owed, most probably, to physical dormancy, with only two exceptions, a population of *L. pedunculatus* with 51 % and another of *L. hispidus* with a percentage of 64 %, both when seeds of age 0 were used.

The highest percentages of germination in seed of age 0 are given by the annual taxa *L. castellanus* and *L. hispidus*, with 36 % and 35,6 % germination respectively. In perennials, percentages of germination are lower and varies between 11.58 % in *L. glaber* and 22.35 % in *L. corniculatus* subsp. *carpetanus*.

Seeds of age 1 show, in general, a decrease in germination percentage in relation to seeds of age 0, with a decrease in annuals (from 36 % to 16,95 % in *L. castellanus* and from 35.56 % to 7.57 % in *L. hispidus*) higher than in perennials. Seeds of age 2 of perennial taxa show an increase in germination percentage in relation to seeds of age 1, while in annual taxa either increase (up to 5.95 % in *L. castellanus*) or is maintained (in *L. hispidus*).

Germination dynamics of seeds of the three ages is shown in table 3. Mean values of t_0 are in general very low in age 0, always less than 7 days; 1.50 to 6.00 days in perennials and 2-3 days in annuals. In seeds of age 1, t_0 values are higher, specially in annual taxa, where t_0 value increase to 5.71 days in *L. hispidus* and to 8,84 in *L. castellanus*. Values of t_0 in seeds of age 2 are always higher than in seeds of age 1, without significant differences between annuals and perennials.

In relation to t_{50} values, both annuals and perennials show a general decrease with age, more pronounced in the former than in the later.

Table 3. Germination dynamics. t_0, time of first germination, in days; t_{50}, time to complete 50 % germination of the total germinated seeds; N, number of populations; $\overline{X}$, mean; σ, standard deviation; v.i., variation interval.

	Age 0							Age 1							Age 2						
	t_0				t_{50}			t_0				t_{50}			t_0				t_{50}		
Taxon	N	$\overline{X}$	σ	v.i.	$\overline{X}$	σ	v.i.	N	$\overline{X}$	σ	v.i.	$\overline{X}$	σ	v.i.	N	$\overline{X}$	σ	v.i.	$\overline{X}$	σ	v.i.
L. corniculatus subsp. *corniculatus*	12	2,33	2,53	1-10	11,17	11,57	3-45	20	3,30	2,18	1-11	11,45	12,08	3-50	2	2,00	0,00	0	7,00	2,83	5-9
L. corniculatus subsp. *alpinus*	1	4,00			4,00			2	2,00	1,41	1-3	7,00	1,41	6-8							
L. corniculatus subsp. *carpetanus*	7	6,00	11,50	1-32	13,00	11,83	3-32	10	3,10	1,20	1-4	7,10	3,31	4-15	2	2,50	0,70	2-3	5,00	1,41	4-6
L. corniculatus subsp. *glacialis*	7	1,56	1,30	1-4	5,56	10,48	1-33	2	2,50	2,12	1-4	4,50	3,54	2-7							
L. corniculatus subsp. *delortii*	2	1,50	0,71	1-2	3,50	0,71	3-4	1	3,00	0,00	0	3,00	0,00	0	1	1,00			4,00		
L. corniculatus subsp. *preslii*	4	2,75	1,71	1-5	3,75	0,96	3-5	2	3,50	0,71	3-4	3,50	0,71	3-4	2	2,00	0,00	0	7,00	5,66	3-11
L. glaber	4	2,36	2,42	1-9	6,91	10,69	1-39	20	3,95	2,74	1-13	9,00	6,97	1-34	2	3,00	0,00	0	7,00	2,83	5-9
L. pedunculatus	26	3,15	2,48	1-10	8,00	5,43	1-22	43	4,33	3,39	1-24	7,12	5,31	3-29	2	3,00	0,00	0	3,50	0,70	3-4
L. castellanus	2	2,00	1,41	1-3	30,00	25,46	12-48	11	8,84	8,44	1-26	16,91	8,72	3-28	2	3,50	0,70	3-4	7,50	3,54	5-10
L. hispidus	3	3,00	0,00	0	33,33	24,66	5-50	7	5,71	2,06	4-9	15,29	13,29	4-44	2	3,50	0,70	3-4	4,00	0,00	0

Table 4. Test of Kruskall-Wallis when population is used as variable of agrupation in seeds of age 0. For abbreviations see table 3.

	Germination %	t_0	t_{50}
L. corniculatus subsp. *corniculatus*	+	-	-
L. corniculatus subsp. *carpetanus*	+	-	+
L. corniculatus subsp. *glacialis*	+	-	-
L. corniculatus subsp. *delortii*	+	-	+
L. glaber	+	-	-
L. pedunculatus	+	+	+
L. castellanus	-	-	+

Figs. 2-4 show germination dynamics of the studied taxa at the three different ages. There are clear differences between annuals and perennials in seeds of age 0, with a more homogeneous increase of germination percentage along the time in perennials than in annuals, which achieve the highest germination percentages mainly in two phases separated by a period of time without a significant increase. These differences do not occur, though, in seeds of age 1 and age 2, where germination dynamics of annuals and perennials is rather similar (see figs. 3 and 4).

Table 4 shows the results of test Kruskal-Wallis, when population is taken as group variable. Significant differences amongst populations of each taxon are indicated by (+); (-) means that there are not differences. For germination percentage there are significant differences for each taxon, with the exception of *L. castellanus*, where all populations behave in a similar way.

When taxon is taken as group variable, statistical analysis does not show any significant difference either for total germination percentage or for t_0, which it is, on the contrary, significant for t_{50}.

Discussion

As demonstrated in this paper, seeds of the studied taxa of *Lotus* sect. *Lotus* show a low germination percentage, as characterise "hard seeds" which have impermeability to water inbibition associate to the presence of one or more palisade cell layers in seed-coat (Baskin & Baskin, 1998), as occurs in *Lotus,* which results in physical dormancy.

Seed size of studied taxa is very variable, but in general seeds are smaller in annuals than in perennial taxa, although *L. corniculatus* subsp. *carpetanus* and *L. pedunculatus* have seeds similar in size to those of annuals. Silvertown (1981) indicates that small seeds germinate more quickly and with higher germination percentage than big seeds. However seeds of *L. corniculatus* subsp. *carpetanus* and *L. pedunculatus* have germination processes similar to those of the other perennials studied.

The results of this paper indicate that germination dynamics and total germination percentages are related to life cycle instead to size. Germination percentages in annuals are higher than in perennials and their seed lose viability when aging.

Perennial taxa can live several years in a same spot and their capacity to keep seed viability is an adaptive advantage as seeds stored in the soil seed bank ensure maintenance of the population. In annuals their high seed germination ensures population survival through the production of a high number of plants which will produce new seeds next growing season with some viable seeds also kept in the soil seed bank.

Germination increase of perennial taxa seeds of age 2 may be owed to strophiole weakness which will favour inbibition (Baskin & Baskin 1998). This may occur in natural conditions owed to temperature changes, soil abrasion, fungi attack, etc., which can drow to lost of physical dormancy. The low lost of viability in seeds of perennials with age, lower than in annuals, agrees with the observations by Britton (1973) who indicates that seeds of perennial taxa keep their germination capacity for many years which contributes to the persistence of these taxa in the soil seed bank.

As far as germination is concerned, the time elapsed by seeds to start germination is rather short, which seems to indicate that seeds take the first favourable conditions to germinate.

Primack (1987) indicates that variations in germination period, physiology, seeds size and general plant morphology allows each species to diversify germination requirements to adapt to different habitats. Germination dynamics of the two annual taxa studied, with two periods of marked increase of germination percentage, could be related to the very marked climatic seasonality of the study area (W Mediterranean). This area is rather arid and germination should be expected to occurs during the first important rainfall after seed dispersal. The first phase should coincide with Autumn rainfall, the resting period with Winter, and the second phase of germination increase should be coincident with late Winter or Spring rainfall.

During the following years differences in germination behaviour tends to be similar both in annuals and perennials as a consequence of viability and dormancy lost.

Kruskal-Wallis test indicates significant differences in germination percentage of different populations of a same taxon in seeds of age 0. This seems to agree with Primack (1987) indications, already mentioned, as populations studied develop in areas with different environmental conditions. No differences have been found amongst populatins of *L. castellanus* owed, most probably, to the fact that they grow in rather similar areas as observed during seed gathering

In relation to germination dynamics there are not differences amongst the studied populations. Seeds tend to germinate soon with independence of taxa and population.

To summarize it can be suspected that variation of germination in *Lotus* sect. *Lotus* does not only depend from genetic conditions as differences into annuals and perennials seem to indicate, but that it also keeps a close relationship to the environmental conditions of the area where the different populations grow.

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Appendix

Origin of populations of which seeds have been used for this study. Population number is given in parenthesis. Supraindexes indicate where seeds of age 0 (0), age 1 (1) or age 2 (2) have been used; of those populations without supraindexes only seeds of age 0 have been used. Sp., Spain; Port., Portugal; It., Italy; Fr., France.

Lotus corniculatus* subsp. *corniculatus: (1[1,2]) Sp.: Madrid, Guadarrama, 24 07 00. (2[1]) Sp.: Segovia, Villacastín, 24 07 00. (3[1]) Sp.: Segovia, Villacastín, 24 07 00. (4[1]) Sp.: Ávila, Hija de Dios, 24 07 00. (5[1]) Sp.: Ávila, Santo Domingo de Posadas, 25 07 00. (6[1]) Sp.: Ávila, Santo Domingo de Posadas, 25 07 00. (7[1]) Sp.: Huesca, El Run, 27 07 00. (8[1,2]) Sp.: Salamanca, La Hoya, 16 08 00. (9[1]) Sp.: Ávila, Palacios de Becedas, 16 08 00. (10[1]) Sp.: Ávila, Hoyocasero, 17 08 00. (11[1])Sp.: Ávila, Venta del Obispo, 17 08 00. (12[1]) Sp.: Ávila, Narros del Puerto, 17 08 00. (13[1]) Sp.: Zamora, Palacios de Sanabria, 17 08 00. (14[1]) Sp.: Zamora, Castellano, 18 08 00. (15[1]) Sp.: Orense, Santa Loacia, 20 08 00. (16[1]) Sp.: Granada, Sierra Nevada, Pico Veleta, 02 08 01. (17[1]) Port.: Viseu, Sao Pedro do Sul, 16 08 01. (18[0,1]) Port.: Aveiro, Vilanova de Gaia, 16 08 01. (19[0,1]) Port.: Porto, Santo Tirso, 16 08 01. (20[0,1]) Port.: Braga, Amarelos, 17 08 01. (21) Sp.: Ávila, Hoyos del Espino, 22 07 02. (22) Sp.: Ávila, Pto. de Mengas, 22 07 02. (23) Sp.: Soria, Covadela, 23 07 02. (24) Fr.: Haut Alpes, Les Vigneaus, 19 08 02. (25) Fr.: Haut Alpes, Montgenevre, 19 08 02. (26) It.: Piamonte, Viverone, 21 08 02. (27) It.: Milano, Secugnago, 22 08 02. (28) It.: Apenini Parmesino, Bore 23 08 02. (29) It.: Genova, St. Stefano d´Abeto, 23 08 02.

L. corniculatus* subsp. *alpinus: (30[1]) Sp.: Navarra, Urbasa, 26 07 00. (31[1]) Sp.: Huesca, Pto. Somport, 26 07 00. (32) Fr.: Savoie, Mont Cenis, 20 08 02.

L. corniculatus* subsp. *carpetanus: (33[1]) Sp.: Huesca, Seira, 27 07 00. (34[1]) Sp.: Huesca, Pto. de Portalet, 27 07 00. (35[1]) Sp.: Huesca, Pto. de Monrepos, 28 07 00. (36[1]) Sp.: Huesca, Almudevar, 28 07 00. (37[1]) Sp.: Lugo, Villalba, 19 08 00. (38[1,2]) Sp.: Granada, Capileira, 23 08 00. (39[1,2]) Sp.: Granada, Trevélez, 24 08 00. (40[1]) Port.: Portalegre, Castello Novo, 15 08 01. (41[1]) Port.: Viseu, Mongualde, 16 08 01. (42) Port.: Aveiro, Albergaria, 16 08 01. (43) Port.: Aveiro, Sta. Maria de Feira, 16 08 01. (44) Port.: Braga, Ponte, 17 08 01. (45[1]) Port.: Braga, Esposende, 18 08 01. (46) Sp.: Cáceres, Pasarón de la Vera, 20 07 02. (47) Sp.: Cáceres, Pasarón de la Vera, 21 07 02. (48) It.: Piacenza, Vernasca, 22 08 02

L. corniculatus* subsp. *glacialis: (49) Sp.: Granada, Borreguiles del Monachil, 02 08 01. (50[0,1]) Sp.: Granada, Veleta, 02 08 01. (51) Sp.: Granada, Veleta, 02 08 01. (52[0,1]) Sp.: Granada, Veleta, 02 08 01. (53) Sp.: Granada, Veleta, 02 08 01. (54) Sp.: Granada, Capileira, 02 08 01. (55) Sp.: Granada, Capileira, 02 08 01.

L. corniculatus* subsp. *delortii: (56[1,2]) Sp.: Navarra, Alozagutia, 26 07 00. (57) Sp.: Burgos, Escuderos, 23 07 02. (58) Fr.: Haut Alpes, Belvedere, 18 08 02.

L. corniculatus* subsp. *preslii: (59[1,2]) Sp.: Álava, Salvatierra, 26 07 00. (60[1,2]) Sp.: Huesca, Pto. de Canfranc, 26 07 00. (61) Fr.: Haut Alpes, Brianconnais, 18 08 02. (62) It.: Susa, Orsiera Rocciabre, 19 08 02. (63) It.: Susa, Pto. Moucenicus, 20 08 02. (64) It.: Vall d´Aosta, Pré St. Didier, 21 08 02.

L. glaber: (65[1]) Sp.: Valladolid, Laguna de Duero, 25 07 00. (66[1]) Sp.: Valladolid, Laguna de Duero, 25 07 00. (67[1,2]) Sp.: Palencia, Venta de Baños, 25 07 00. (68[1]) Sp.: Palencia, Torquemada, 25 07 00. (69[1]) Sp.: Burgos, Castañares, 25 07 00. (70[1]) Sp.: Burgos, Quintanapaya, 25 07 00. (71[1]) Sp.: Burgos, Quintanavides, 25 07 00. (72[1]) Sp.: Burgos, Pancorbo, 25 07 00. (73[2]) Sp.: Álava, Vitoria, 26 07 00. (74[1]) Sp.: Navarra, Altsasu, 26 07 00. (75[1]) Sp.: Navarra, Muruarte, 26 07 00. (76[1]) Sp.: Navarra, Zabalegui, 26 07 00. (77[1]) Sp.: Navarra, Yesa, 26 07 00. (78[1]) Sp.: Zaragoza, Calatayud, 28 07 00. (79[1]) Sp.: Zaragoza, 28 07 00. (80[1]) Sp.: Zamora, Zamora, 17 08 00. (81[1]) Sp.: Zamora, Castellanos de Sanabria, 17 08 00. (82[1]) Sp.: Zamora, Anta de Rioconejos, 18 08 00. (83[1]) Sp.: León, Castrocontigo, 18 08 00. (84[1]) Sp.: León, Truchas, 18 08 00. (85[1]) Port.: Portalegre, Castello Novo, 15 08 01. (86) Port.: Coimbra, Coimbra, 18 08 01. (87) Sp.: Burgos, Mortigüela, 23 07 02. (88) Sp.: Burgos, Castillo de la Reina, 23 07 02. (89) Sp.: Burgos, Palacios de la Sierra, 23 07 02. (90) Fr.: Aubres, 18 08 02. (91) Fr.: Aubres, Belleconde, 18 08 02. (92) Fr.: Verclause, Nyons, 18 08 02. (93) Fr.: Haut Alpes, Gap, 18 08 02. (94) Fr.: Haut Alpes, Belvedere, 18 08 02. (95) It.: Piacenza, Fiorenzuola d´ Arda, 22 08 02. (96) It.: Piacenza, Vernasca, 22 08 02.

L. pendunculatus: (97[1]) Sp.: Huelva, Aracena, 22 06 00. (98[1]) Port.: Faro, Carrapateira, 13 07 00. (99[1,2]) Port.: Bejas, Odemira, 13 07 00. (100[1]) Port.: Bejas, Milfontes, 13 07 00. (101[1]) Port.: Bejas, Milfontes, 13 07 00. (102[1]) Port.: Setúbal, Santiago do Cacen, 14 07 00. (103[1]) Sp.: Ávila, Hija de Dios, 24 07 00. (104[1]) Sp.: Burgos, Arlanzón, 25 07 00. (105[1]) Sp.: Cáceres, Plasencia, 16 08 00. (106[1]) Sp.: Cáceres, Zarza de Granadilla, 16 08 00. (107[1]) Sp.: Cáceres, Herbás, 16 08 00. (108[1]) Sp.: Cáceres, Baños de Montemayor, 16 08 00. (109[1]) Sp.: Cáceres, Baños de Montemayor, 16 08 00. (110[1]) Sp.: Salamanca, Cantagallo, 16 08 00. (111[1]) Sp.: Salamanca, La Hoya, 16 08 00. (112[1]) Sp.: Ávila, Aliseda de Tormes, 16 08 00. (113[1]) Sp.: Ávila, Navacepeda de Tormes, 16 08 00. (114[1]) Sp.: Zamora, Otero de Sanabria, 17 08 00. (115[1]) Sp.: Zamora, Trefacio, 18 08 00. (116[1]) Sp.: Zamora, Anta de Rioconejos, 18 08 00. (117[1]) Sp.: León, Ambasmestas, 18 08 00. (118[1]) Sp.: León, Quintela, 18 08 00. (119[1]) Sp.: Lugo, Cervantes, 18 08 00. (120[1,2]) Sp.: Lugo, Penamil, 18 08 00. (121[1]) Sp.: Lugo, Fonsagrada, 19 08 00. (122[1]) Sp.: Lugo, Paradabella, 19 08 00. (123[1]) Sp.: Lugo, Meira, 19 08 00. (124[1]) Sp.: Lugo, Balmonte, 19 08 00. (125[1]) Sp.: La Coruña, Picaraña, 20 08 00. (126[1]) Sp.: La Coruña, Taragona, 20 08 00. (127[1]) Sp.: Pontevedra, Pontevedra, 20 08 00. (128[1]) Sp.: Granada, Capileira, 23 08 00. (129[1]) Sp.: Granada, Trevélez, 24 08 00. (130[1]) Sp.: Granada, 24 08 00. (131[0,1]) Sp.: Granada, Veleta, 02 08 01. (132) Sp.: Granada, Capileira, 02 08 01. (133) Sp.: Granada, Pampaneira, 04 08 01. (134[1]) Sp.: Granada, Pampaneira, 04 08 01. (135[0,1]) Sp.: Granada, Busquistas, 04 08 01. (136) Port.: Portalegre, Reguengo, 14 08 01. (137[0,1]) Port.: Portalegre, Povoas e Meadas, 14 08 01. (138[0,1]) Port.: Portalegre, Nisa, 14 08 01. (139) Port.: Portalegre, Castello Novo, 15 08 01. (140) Port.: Portalegre, Castello Branco, 15 08 01. (141) Port.: Portalegre, Penchas de Saude, 15 08 01. (142[0,1]) Port.: Portalegre, Manteigas, 15 08 01. (143[0,1]) Port.: Portalegre, Gouveia, 16 08 01. (144) Port.: Viseu, Viseu, 16 08 01. (145) Port.: Braga, Viera do Minho, 17 08 01. (146[0,1]) Port.: Braga, Caldas do Geres, 17 08 01. (147[0,1]) Port.: Viana do Castelo, Vila Nova de Carreira, 18 08 01. (148) Sp.: Cáceres, Garganta Olla, 20 07 02. (149) Sp.: Cáceres, Arroyomolino, 21 07 02. (150) Sp.: Cáceres, Arroyomolino, 21 07 02. (151) Sp.: Ávila, La Nava del Barco, 21 07 02. (152) Sp.: Ávila, Angostura, 22 07 02. (153) Sp.: Ávila, San Juan de Gredos, 22 07 02. (154) Sp.: Ávila, Pto. de Mengas, 22 07 02. (155) Sp.: Toledo, Navalucillos, 24 07 02. (156) Sp.: Toledo, Robledo del Buey, 24 07 02. (157) Sp.: Cáceres, Guadalupe, 24 07 02.

L. castellanus: (158[1]) Sp.: Cádiz, Pto. Sta. María, 13 06 00. (159[1]) Sp.: Huelva, Almonaster, 28 06 00. (160[1,2]) Sp.: Huelva, Almonaster, 28 06 00. (161[1]) Sp.: Huelva, El Repilado, 28 06 00. (162[1]) Sp.: Cádiz, Alcalá de los Gazules, 06 07 00. (163[1]) Port.: Bejas, Cercal, 13 07 00. (164[1]) Sp.: Huelva, Puebla de Guzmán, 14 07 00. (165[1,2]) Sp.: Madrid, Galapagar, 24 07 00. (166[1]) Sp.: Málaga, Cortes de la Ftra., 04 08 00. (167) Port.: Portalegre, Vila Velha de Rodao, 15 08 01. (168[0,1]) Port.: Leiria, Leiria, 18 08 01. (169[1]) Port.: Santarem, Fatima, 18 08 01.

L hispidus: (170[1]) Sp.: Sevilla, Castillo de las Guardas, 22 06 00. (171[1,2]) Port.: Faro, Carrapateira, 13 07 00. (172[1]) Port.: Setúbal Santiago do Cacen, 14 07 00. (173[1,2]) Sp.: Huelva, Rosal de la Ftra., 14 07 00. (174[1]) Sp.: Cáceres, Baños de Montemayor, 16 08 00. (175[1]) Sp.: Pontevedra, Carracedo, 20 08 00. (176[1]) Sp.: Pontevedra, Pontevedra, 20 08 00. (177) Port.: Portalegre, Vila Velha de Rodao, 15 08 01. (178) Port.: Braga, Ponte, 17 08 01. (179) Port.: Braga, Ponte, 17 08 01.