VARIATION IN THE GRASS PEA (*LATHYRUS* SATIVUS L.) AND WILD SPECIES

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VDEX WORDS

Lathyrus sativus, grass pea, wild species, variation, multivariate analyses.

SUMMARY

Forty-nine accessions of *Lathyrus sativus* were studied for morphological variation. Data were analysed using Principal Components Analysis and Cluster Analysis. The variation in 14 species of Section *Lathyrus* was also evaluated in order to ascertain affinities between *L. sativus* and other species.

L. sativus is a highly variable species, and there is a clear distinction between the blue-flowered forms from south-west Asia, Ethiopia and the Indian subcontinent, and the white and white and blue flowered forms with white seeds which have a more westerly distribution. Differences in vegetative parts may be due to selection for forage types.

L. sativus appears to be closely related to L. cicera and L. gorgoni, and this relationship needs further investigation.

INTRODUCTION

The genus *Lathyrus* is large with 187 species and subspecies recognised (ALLKIN et al., 1983). Species are found in the Old World and the New World, but clearly there are centres of diversity for Old World species in Asia Minor and the Mediterranean region (Zeven & De Wet, 1982). A number of species are used as animal fodder plants including L. hirsutus and L. palustris, and some are valued as ornamentals, especially L. odoratus, the sweet pea. Only one species, L. sativus, the grass pea, khesari or chickling pea is widely cultivated as a food crop, and this pulse is a dependable cropper in drought conditions (SMARTT, 1976). It is grown to the greatest extent in India, and to a lesser degree in the Near East and the Mediterranean region. The crop grows well on almost all types of soil and is suited to dry climates, but can apparently tolerate water-logging (PURSEGLOVE, 1968; DUKE et al., 1981). The drawback to the use of this pulse is the production of a neurological disorder called lathyrism in man and domestic animals when the seeds are consumed in excess, especially when other food crops are scarce in times of drought. Lathyrism causes a non-reversible paralysis of the lower limbs in man, the causes of which are uncertain but which appears to be due to the presence of a high concentration of an uncommon amino acid, α -amino- β

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oxalylamino propionic acid (BELL, 1964). Undoubtedly the genus is an important one. and L. sativus is an ancient cultigen. Its origin, however, is unknown (BALL, 1968), but it is considered to be native to southern Europe and south-west Asia (DUKE et al., 1981). There have been reports of wild L. sativus in Iraq (TOWNSEND & GUEST, 1974) but whether these plants are truly wild or escapes from cultivation has not yet been clarified. Some of the earliest archaeological evidence comes from Jarmo, in Iraqi Kurdistan, dated at 8000BP (HELBAEK, 1965). Remains of Lathvrus species have been found at Ali Kosh (9500-7600BP) and Tepe Sabz (7500-5700BP) in Iran, and are amongst the most common foods recorded for these sites. At Azmaska Moghila, in Bulgaria, remains dated at c. 7000BP, have been identified tentatively as L. cicera (RENFREW, 1969). SARASWAT (1980) gave a later date (4000-3500BP) for the cultivation of L. stativus in India, and indicated the possibilities of diffusion of the crop from West Asia, and remains have been dated at 3800-3200BP from Navdatoli (ALLCHIN, 1969). VAVILOV (1951) described two separate centres of origin of the crop. One was the Central Asiatic Centre which includes north-west India (the Punjab, the northwestern frontier provinces and Kashmir), Afghanistan, the Soviet Republics of Tadjikstan and Uzbekistan, and western Tian-Shan. The second was the Abyssinian Centre. In addition, Vavilov noted trends in diversity similar to those found in other pulses, such as lentils, in that smaller seeded forms were found in southern and south-west Asia, whereas around the Mediterranean region, almost all were highly cultivated forms with large white seeds and flowers.

Recent cytogenetical and biosystematic studies on other pulses such as lentils (ZOH-ARY, 1972) and chickpeas (LADIZINSKY & ADLER, 1976) have focussed attention on wild species genetic resources, and their more efficient utilization in plant breeding. In this paper, we have evaluated the morphological variation in L. sativus using living material, in order to determine the extent of diversity in the species. In addition a herbarium survey of L. sativus and wild species was made with the objective of identifying wild species more closely related to the cultigen.

MATERIALS AND METHODS

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Variation in L. sativus. Forty-nine accessions of L. sativus were evaluated in this study, as listed in Table 1. They were planted in a randomised complete block with five replicates, each accession being represented by three plants per replicate. Measurements were only made on the middle plant of each group of three from each accession. The seeds were planted in early May 1982, and after three weeks, were transplanted to the ground area in a large glasshouse at a planting distance of 0.45 m between rows, and 0.15 m between plants. Two weeks after sowing, all plants were inoculated using a culture of *Rhizobium leguminosarum*, supplied by Rothamsted Experimental Station, Harpenden. Seven weeks after sowing, the plants started to flower, and the date for each accession was recorded. Some 16 quantitative characters were measured, and four qualitative characters assessed, as listed in Table 2. Four months after sowing, pods and seeds were harvested from mature plants.

Herbarium survey. Herbarium material from the Royal Botanic Gardens, Kew, was assessed for variation in *Lathyrus* section *Lathyrus* (KUPICHA, 1974). Some 14 species which were previously classified in Section Cicercula were studied, and twelve morpho-

B'ham acc. no.	Ргочепансе	Institute	Acc. No.
0008	Portugal	Oeiras	487
0017	Portugal	Oeiras	486
0043	Canary Isles (Fuerteventura)	Genus	400
0428	Turkey	Izmir	39180
0429	Turkey	Izmir	35048
0430	Ethiopia	Vavilov	WIR-1321
0432	Portugal	Vavilov	WIR-1368
0433	Hungary	Vavilov	WIR-1474
0434	Hungary	Vavilov	WIR-1336
0435	Soviet Union (Tambor Region)	Vavilov	WIR-1247
0438	India	Vavilov	WIR-1309
0439	India	Vavilov	WIR-1315
0441	India	Vavilov	WIR-1311
0442	Soviet Union (Voronezh Region)	Vavilov	WIR-21
0443	Soviet Union (Ukrainian SSR)	Vavilov	WIR-1341
0444	Soviet Union (Georgian SSR)	Vavilov	WIR-965
0446	Soviet Union (Krasnodar Territory)	Vavilov	WIR-1307
0447	Bulgaria	Vavilov	WIR-1297
0448	Soviet Union (Tajik SSR)	Vavilov	W1R-980
0452	Soviet Union (Azerbaijan SSR)	Vavilov	WIR-1179
0454	Turkey	Vavilov	WIR-864
0455	Afghanistan	Vavilov	WIR-1487
0459	Soviet Union (Mordovian SSR)	Vavilov	WIR-[298
0466	Soviet Union (Azerbaijan SSR)	Vavilov	WIR-1215
0467	Soviet Union (Orenburg Region)	Vavilov	WIR-1190
0472	Soviet Union (Moldavian SSR)	Vavilov	WIR-1512
0473	Bulgaria	Vavilov	WIR-1364
0474	Soviet Union (Tatar SSR)	Vavilov	WIR-1211
0475	Soviet Union (Tambor Region)	Vavilov	WIR-1248
0476	Turkey	Vavilov	WIR-1363
0479	Soviet Union (Georgian SSR)	Vavilov	WIR-968
0480	Soviet Union (Tajik SSR)	Vavilov	WIR-1165
0481	Soviet Union (Ukraiman SSR)	Vavilov	WIR-1330
0482	Algeria	Vavilov	WIR-1486
0483	Hungary	Vavilov	WIR-1505
0484	Soviet Union (Krasnodar Territory)	Vavilov	WIR-1253
0485	Soviet Union (Kuibyshev Region)	Vavilov	WIR-1246
0486	Crete	Vavilov	WIR-1508
0488	Bulgaria	Vavilov	WIR-1451
0489	Czechoslovakia	Vavilov	WIR-1450
0491	India	Vavilov	WIR-1445
0492	Czechoslovakia	Vavilov	WIR-1500
0493	Ethiopia	Vavilov	WIR-1320
0494	Soviet Union (Azerbaijan SSR)	Vavilov	WIR-1189
0495	Soviet Union (Orel Region)	Vavilov	WIR-1469
0496	Portugal	Vavilov	WIR-1440
0498	Soviet Union (Kuibyshev Region)	Vavilov	WIR-1522
0502	Soviet Union (Krasnodar Territory)	Vavilov	WIR-1392
0506	Afghanistan	Vavilov	WIR-1444

Table 1. Lathyrus sativus accessions evaluated in this study.

Luph: nea 33 (1984)

Character	Field Trial	Herbarium Survey
Leaf length	+	+
Leaflet length	+	+ +
Leaftet breadth	+	+
Shape of leaflet		+
Petiole length	+	+
Peduncle length	+	+
No. flowers/peduncle		+
Arista length	+	
Standard petal length	+	
Standard petal breadth	+	
Length of flower		+
Calyx length	+	+
Calyx teeth length	+	+
Style length	+	
Style breadth	+	
Stipule length	+	+
Stipule breadth	+	+
Pod length	+	
Pod breadth	+	
Flower colour	+	
Seed colour	+	
Days to flowering	+	

Table 2. Characters evaluated for variation in *L. sativus*, and in the herbarium survey of related species in section *Lathyrus*.

logical characters (Table 2) were scored on 121 specimens.

Numerical analyses. Analyses of variance were carried out on all quantitative data. The data were also analysed by Principal Components Analysis and Cluster Analysis (Euclidean Distance plus Ward's Method), using the Clustan 1C Computer package (WISHART, 1978). Character correlations were also calculated.

RESULTS

Simple statistics for the quantitative characters are shown in Table 3. Most of the characters show a high variation, except standard petal length and breadth, and style and pod length, which all have a coefficient of variation of less than 10%. Vegetative characters such as leaf and leaflet length were much more variable. For all but five quantitative characters, namely leaf length, leaflet length, peduncle length, corolla length, and stipule length, variation between accessions was greater than within accessions (Table 4).

Flower colours of three types were observed: blue, white, and white with some blue pigmentation on the standard petal, the lateral petals or both. None of the accessions studied had pink flowers, although such types have been reported elsewhere (VAVILOV, 1951; KAUL et al., 1982). Seed colour was variable, either plain or speckled, and the seed main colour was white, light brown or grey. Many of the character correlations

Character	Mean	Coefficient of variation (%)	Range
Leaf length	99.34	20.6	47.0-194.0
Leaflet length	67.42	22.7	32.0-150.0
Leaflet breadth	6.55	33.6	2.0-15.7
Petiole length	31.14	21.1	15.0-49.0
Peduncle length	42.58	29.4	18.0- 80.0
Arista length	1.69	48.5	0.1- 4.0
Standard petal length	14.78	8.9	4.1~ 17.9
Standard petal breath	10.98	8.6	6.0-13.7
Calyx length	10.83	11.0	7.0-17.3
Calyx teeth length	6.91	15.5	0.7- 9.9
Style length	6.36	8.3	4.9-10.3
Style breadth	0.75	18.7	0.4-1.1
Stipule length	24.62	20.0	13.0- 40.5
Stipule breadth	4.09	25.0	2.0- 7.8
Pod length	39.14	8.7	30.0- 50.0
Pod breadth	13.73	16.7	6.0-33.0
Days to flowering	49.98	6.0	41.0- 53.0

Table 3. Simple statistics of quantitative characters evaluated in L. sativus accessions. Measurements in mm.

Table 4. Variances of 17 quantitative characters studied in 49 accessions of Lathyrus sativus.

	Between	Within	
	accessions	accessions	
	(48 df)	(192 df)	
Leaflength	447.396	374.813	
Leaflet length	245.995	211.947	
Leaflet breadth	7.506	4.034	
Petiole length	74.713	33.416	
Peduncle length	169.540	142.385	
Arista length	0.986	0.584	
Standard petal length	1.898	1.669	
Standard petal breadth	1.564	0.714	
Calyx length	2.004	1.170	
Calyx teeth length	1.699	0.953	
Style length	0.530	0.218	
Style breadth	0.041	0.014	
Stipule length	27.918	23.363	
Stipule breadth	1.416	1.007	
Pod length	21.216	9.459	
Pod breadth	6.844	2.190	
Days to flowering	16.190	6.590	
Days to nowering	10.190	0.390	

were as expected, with correlations in size between the different plant parts (Table 5). Nevertheless, some other correlations such as leaflet length and peduncle length are interesting, as are the correlations of various leaf characters with stipule characters. The correlations of flower and seed colours were very high, with white-flowered accessions having only white seeds, with no secondary markings on the testa. From these correlations it is apparent that the accessions with the broadest pods, and by inference also with the largest seeds, had white or white and blue flowers and white seeds, where-as blue-flowered accessions had smaller, coloured seeds with secondary markings on the testa.

Principal Components Analysis. The first six eigenvectors accounted for almost 80% of the variation, with the first two accounting for 30.97% and 15.60% respectively. On Component 1, the accessions were distributed on the basis of the size of plant parts, with stipule length best describing the variation pattern, but with significant contributions from leaf length and petiole length. The accessions were distributed on Component 2 on the basis of seed colour, but flower colour also had an almost equal variance of -0.452, compared to -0.467. The results are presented in Fig. 1. Thus there

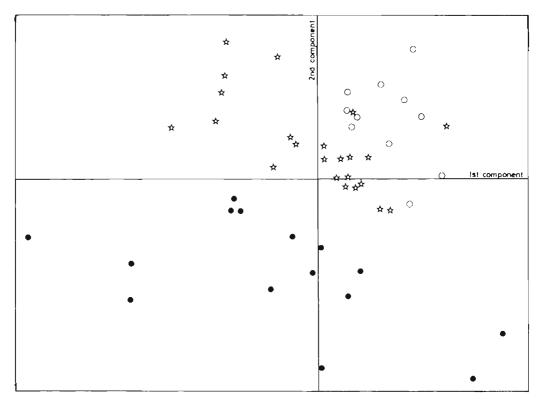


Fig. 1. Diagram expressing morphological variation of 49 accessions of *L. sativus*, based on mean values, obtained from the first two components of Principal Components Analysis (\bigcirc = white flowers; a = white and blue flowers; $\textcircled{\bullet}$ = blue flowers).

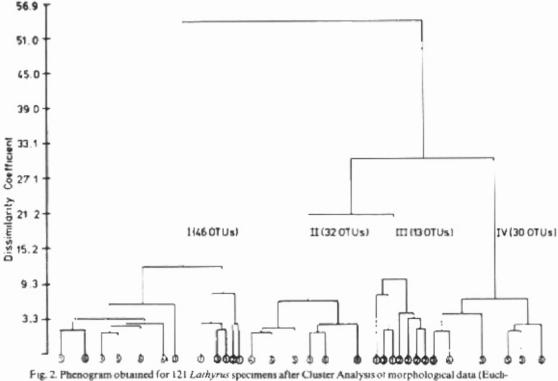
Character	2	3	4	5	6	7	8	9	10	П	12	13	14	15	16	17	18
1. Leaf length	0.91	0.45	0.76	0.50							0.73	0.52					
2. Leaflet length		0.49	0.47	0.51							0.60	0.41					
3. Leaflet breadth				0.46								0.57					
4. Petiole length											0.67	0.52	0.53				
5. Peduncle length								0.45	0.41		0.44	0.49			0.41		
6. Standard petal length						0.70											
7. Standard petal breadth																	
8. Calyx length								0.86	0.53			0.44	0.41				
9. Calyx teeth length									0.54		0.47	0.44					
10. Style length										0.46		0.51					
11. Style breadth																	
12. Stipule length												0.65	0.44		0.47		
13. Stipule breadth													0.44		0.42		
14. Pod length														0.57			
15. Pod breadth																-0.59	-0.61
16. Days to flowering																	
17. Flower colour																	1.0
18. Seed colour																	

Table 5. Correlations among various characters in 49 accessions of L. sativus, based on mean values. Only values greater than r \pm 0.4 are shown (all values significant at p = 0.01).

is a clear separation of the *L. sativus* accessions into two main groups. One group comprises the blue-flowered forms quite distinct from the rest, whereas the other group has both white and mixed flower colour types. Within each of these groups, there is a distribution from small to large stipules and other vegetative parts, as would be expected on the basis of the significant correlations between these characters.

Geographical distribution. The 49 accessions were selected from a much larger collection of *L. sativus* accessions, so as to cover the known geographical distribution of the species. The blue-flowered varieties all came from south-west and south Asia, and Ethiopia, whereas the white and mixed coloured varieties had a more western distribution, from the Canary Isles to the western republics of the Soviet Union.

Herbarum survey. Two major groups were formed by the Cluster Analysis, at a dissimilarity coefficient of 54.2, with 46 and 75 OTU's respectively, as illustrated in Fig. 2. The larger of the groups further divided at a coefficient of 30.9, with 45 and 30 OTU's respectively. A further subdivision of this group of 45 occurred at a coefficient of 21.2 to form two groups with 32 and 13 OTU's each. Below this level of dissimilarity there are many further subdivisions, but many of these occur at more or less the same level. Consequently four main groups can be recognised, and the species composition of each is given in Table 6. It is interesting to note that all L, sativus specimens were placed in Group 1, as were the majority of L. cicera and L, gorgoni specimens, the



dean Distance plus Ward's method)

Species	Specimens	Grou	p .		÷
	examined	1	2	3	4
L. annuus	. 25	1	20	3	• 1
L. blepharicarpus	9	-	~	-	9
L. cassius	6		1	-	5
L. chloranthus	1	2	-	-	1
L. chrysanthus	2	_	÷ -	2	-
L. cicera	21	16	, - <u>-</u>	-	5
L. gorgoni	12	11	~	-	, 1
L. hierosolymitanus	4	-	4	-	
L. hirsutus	17		7	4	6
L. lycicus	2		- '	2	-
L. marmoratus	2	-	. –	- ,	2
L. sativus	16	16	1.15	,	_
L. stenophyllus	2	. 2	3.5	-	-
L. trachycarpus	2	-	5.02	2	-
Total	121	46	32	13	30

Table 6. Species composition of groups formed by Cluster Analysis (Euclidean Distance plus Ward's method).

two specimens of *L. stenophyllus*, and one of *L. annuus*. Group 4 consisted of all *L. blepharicarpus* specimens as well as a few specimens of species such as *L. cicera*, *L. gorgoni* and *L. hirsutus*, the majority of which had been placed in other groups.

DISCUSSION

Through the application of multivariate methods, it has been possible to analyse and describe the variation in L. sativus and to assess the relationships of L. sativus and some species in Section Lathyrus.

L. sativus shows great morphological variation, especially in vegetative characters such as leaf length, while floral characters are much less variable. This array of variation is undoubtedly the result of geographical separation as well as selection by man. The pattern of variation in L. sativus based on flower colour and seed coat colour agrees broadly with that described by VAVILOV (1951), who also noted that forms of L. sativus with white flowers and white seeds were highly selected and recessive. Such forms were typical of a more western distribution around the Mediterranean Basin. One can postulate that the forms with blue flowers and speckled seeds are more primitive, and the pattern of variation found in L. sativus is not unlike that found in other pulses such as lentils and broad beans. In these two crops, primitive forms with small seeds have a distribution to the east of the Mediterranean (Turkey, Transcaucasia, Afghanistan), whilst larger seeded forms have been selected in the Mediterranean region. Seed size was not evaluated in this study due to the small samples available. Nevertheless one can infer that some accessions were small-seeded based on pod dimensions.

The development of forms with larger leaves may have resulted from selection for forage types. Certainly the grass pea is consumed as a pulse in India, Bangladesh,

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and in other countries of south-west Asia, whilst in the Mediterranean region it appears to be more of a forage plant. This does not however explain differences in seed size among grass pea varieties.

The close morphological affinity of L. sativus, L. cicera and L. gorgoni is interesting, and may be a consequence of hybridisation or common ancestry. L. sativus and L. cicera resemble each other in certain floral characteristics, although in fruit, L. sativus is closer to L. amphicarpos, L. blepharicarpus and L. marmoratus (DAVIS, 1970). The relationship of L. stenophyllus to these three species is more problematical on account of the small number of specimens examined. From the material studied, there was a clear distinction between these three species and the other 11 species of section Lathyrus, with a few exceptions. In addition, the pollen morphology of L. sativus, L. cicera and L. gorgoni is very similar (YUNUS, 1982), and preliminary karyotype studies of these three species show close similarity. DAVIES (1958) has indicated a closer link between L. sativus and L. cicera than to other species. Close association of L. sativus with L. cicera and L. gorgoni has also been shown by BELL (1971), on the basis of the distribution of non-protein amino acids in Lathyrus species. It is interesting to note that although L. hirsutus had originally been classified with these three species in Section Cicercula, its pollen morphology was very distinct, and much closer to other species far removed from L. sativus and its allies (YUNUS, 1982).

In general, species hybrids are difficult to make in Lathyrus (SENN, 1938), but the reasons for this interspecific incompatibility are not at all evident. The ability of L. sativus and L. cicera to hybridise was demonstrated by SAW LWIN (1956) suggesting a closer relationship between these two than to other species. In general, L. sativus appears to be autogamous (from observations on glasshouse plants), although in nature the possibility of some outcrossing cannot be discounted, and which could lead to interspecific hybridisation. Certainly, L. sativus, L. cicera and L. gorgoni have a sympatric distribution in Turkey, and are often found as weeds of corn fields. An evaluation of species gene-pools (HARLAN & DE WET, 1971) is a valuable step in the exploitation of germplasm (SMARTT, 1981), and this approach has been applied successfully in the elucidation of chickpea evolution (LADIZINSKY & ADLER, 1976). Such an approach may prove worthwhile in Lathyrus breeding if it is necessary to reach outside the cultivated species gene-pool for valuable genetic traits. Undoubtedly the grass pea is a pulse with considerable potential for the future, but the problem of lathyrism restricts its exploitation. There is apparently some variation in the content of some of the neurotoxins (KAUL et al., 1982), and progress is being made in the Indian subcontinent in the selection of less toxic lines. Nevertheless, further exploitation of L. sativus germplasm and the development of the crop will only be achieved once this risk is eliminated. The origin and evolutionary history of L. sativus still remain a mystery, although there is now some evidence to support further research with this pulse.

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