ENVIRONMENTAL AND GENOTYPIC EFFECTS ON POD CHARACTERISTICS RELATED TO COMMON BEAN QUALITY

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One figure, four tables

Abstract

A collection of 121 common bean (*Phaseolus vulgaris* L.) landraces from Spain and Portugal (Iberian Peninsula) was evaluated in six environments (three locations and two years). Significant differences among landraces were found for period of flowering, pod maturity, and pod morphology (weight, length, width/thickness and curvature). Wide variation among landraces was revealed by the range of variation observed. Environmental effects were not significant although year x location and landrace x year x location interactions were significant for all traits studied except for pod curvature. Poor consistent expression across the different environments for most of the traits studied was expressed by the low values of constancy (C) with the exception of width/thickness indicating that pod shape (round or flat) was expressed regularly across different environments. Principal component analysis enhanced differences among environment showed that 51 landraces were adapted to specific environments and only four of them had broad geographic adaptability with similar performance under different conditions. These results could be a starting point for selection of new inbred lines adapted to distinct environments with potential for the improvement of current snap bean cultivars.

Key words: *Phaseolus vulgaris*, breeding, landraces, snap bean, variance components, environment interaction

Running title: Bean pod quality

Introduction

Common bean (*Phaseolus vulgaris* L.) landraces are frequently grown together with other crops in small farms in Spain and Portugal for self consumption and also to be sold in local markets. New commercial cultivars are displacing landraces but a high degree of diversity is still maintained within this species (De Ron et al. 1997; Rodiño et al. 2001) and farmers often use bean landraces both as vegetable (fresh pods) and as grain legume (dry seed). Advantages for small farmers are clear since they get two different on-farm supplies from one crop.

Snap beans are a type of common bean grown for fresh market consumption selected for tasty pods with reduced fiber. Important pod characteristics include length, cross-section shape, sieve size, color, smoothness, fiber and seed index (Silbernagel and Drake 1978).

Breeders look for snap bean lines well adapted to each region but snap beans represent an almost closed population with low genetic variability. In the genetic regions subject to active selection, reduction of diversity occurred leading to a reduction of the information given by microsatellites (Métais et al. 2002). This could be regarded as a success of the selection process but it introduces an asymptote to the genetic progres. There is a general need for the identification and transfer of traits from other backgrounds into snap beans, thus dry beans may be used as a source of genetic variability.

Much of the genetic improvement of snap bean has been achieved through the selection of varieties by applying conventional breeding techniques of self-pollinated crops (Silbernagel et al. 1991; Sills and Nienhuis 1993). There is available information about bean germplasm evaluation and characterization in different areas that sometimes includes dry and snap beans (Silbernagel and Drake 1978; Escribano et al. 1990; Gil and De Ron 1992; Mullins et al. 1999; Piergiovani et al. 2000; Fourie 2002).

Environmental effects and interactions can affect quantitative and complex traits but qualitative characters controlled by a few genes are expected to be environmentally insensitive. A reliable estimation of the variation in characters affecting pod quality should be useful for snap bean breeders. The objectives of this research were: i) to describe the variation in agronomic and pod quality traits in common bean landraces, ii) to estimate the environmental effects and interactions in these traits and iii) to assess the use of the quantitative variation described in breeding for snap bean cultivars.

Material and Methods

One hundred and twenty one common bean landraces from Spain and Portugal were evaluated during the spring-summer season in 1993 and 1994 in three locations in Spain: Pontevedra (42° 26' N, 8° 38' W, 40 masl, average temperature 14.6 °C, average annual rainfall 1600 mm), Lalín (42° 36' N, 8° 8' W, 500 masl, average temperature 11.7 °C, average annual rainfall 1200 mm) and Vitoria (42° 51' N, 2° 40' W, 530 masl, average temperature 11.7 °C, average annual rainfall 1200 mm). These landraces were a part of the germplasm collection at the Misión Biológica de Galicia (MBG-CSIC, Pontevedra, Spain) (De Ron et al. 1997).

Field trials were arranged according to randomized complete blocks with two replications. Each accession was sown in a single 15 plant-row plot with row to row distance of 0.80 m and plant to plant distance of 0.25 m, equivalent to a crop density of 50000 plants/ha.

During the growing period, data were taken on quantitative traits regarding to plant and pod

(Puerta-Romero 1961; IBPGR 1982; CIAT 1984; De la Cuadra et al. 2001): flowering duration (days from 50% of plants showing open flowers until all plants had not any flower), fresh pod maturity (days from sowing until 50 % of plants present fresh pods at commercial stage), pod data (averaged from five immature pods): length (linear distance in millimetres from the pod apex to the top), straight length (distance in millimetres from the pod apex to the top), width (distance in millimetres at right angles to the sutures at the level of the second seed from the apex), width/thickness ratio or coefficient K (Puerta-Romero 1961) (being thickness the distance between pod sides at the level of the second and third seed from the apex), this index gives information of the cross-section shape (round or flat) of the immature pod, weight (expressed in grams and determined when pods reached the commercial stage for fresh consumption), and curvature was expressed as the ratio length/straight length. Additionally, growth habit (CIAT 1984), pod fiber (scored from 1-low to 5-high), and pod colour were determined in all the accessions evaluated.

Analyses of variance (SAS Institute 2000) were performed for all the quantitative traits studied over the 121 landraces, for the two years and three locations. From the combined analysis of variance it was estimated the ratio genetic versus non-genetic variance or constancy (C) (Goodman and Paterniani 1969; De Ron and Ordás 1989). Error was calculated according to Comstock and Moll (1963) and Hallauer and Miranda Fo. (1981). Principal component analyses displaying the ordination of landraces in the different environments were made by means of the NTSYS-pc package (Rohlf 2000).

Results

Table 1 shows the variation of the 121 landraces for growth habit, pod color and pod fiber. Pod fiber and growth habit displayed wide variation and the majority of landraces had green pod color. Variation among the landraces regarding to the quantitative traits evaluated is displayed in Table 2, by means of a combined analysis of variance over years and locations, means and range of variation. Differences among landraces were significant for the six traits evaluated. Year x location and landraces x year x location interactions were significant for all the traits with the exception of pod curvature. Since the studied landraces are a wide sample of germplasm, ranges of variations were also wide. The components and ratio of variance derived from the analysis of variance are shown in Table 3.

Table 4 displays the variation across the six environments on five quantitative traits which showed interactions with years and locations. There is a general trend towards a combined

effect of year and location on the phenotype of plants regarding to these five traits.

Figure 1 represents the distribution of landraces in each one of the six environments according to the first (PC 1) and second (PC 2) or third (PC 3) principal components that accounted from 67.7 % to 83.2 % of the variation. Pod size variation is represented by PC 1 while PC 2 or PC 3 explains earliness. Distinct distribution of landraces in each environment could be interpreted as clear adaptation of landraces to specific environmental conditions. The arrows in the six plots mark the quarter where the most valuable landraces were ordinated according to earliness and pod size in each environment. The majority of them (80 %) appeared in the marked area in one, two or three of the studied environments, some other in four or five (20 %) and none landrace is present in the marked quarters in all the environments.

Discussion

High variation for qualitative traits among the 121 landraces studied was shown in the results of the field trials across the six environments (Table 1). Most common growth habit was indeterminate climbing type, which agreed with previous reports of evaluation of Spanish germplasm (Gil and De Ron 1992). Intercropping with maize is still common in the North of Spain and Portugal (Santalla et al. 1994; Santalla et al. 1999) and indeterminate climbing type is the most appropriate plant architecture for this cropping system. The majority of the landraces are supposed to be used as dry bean that explains the high to medium presence of fiber and the green and green-purple color of the pods. The presence of 39 landraces with low fiber in the pod and six yellow-podded landraces are important since these phenotypes should be favourable for selection of snap bean inbred lines.

Significant landrace differences were present in all the quantitative traits (Table 2) that agrees with previous studies by Natarajan and Arumugan (1979), Joshi and Mehra (1984) and Gil and De Ron (1992). No significant environmental effects were detected for years and locations while interactions year x location and landraces x year x location were significant in all the traits evaluated except for pod curvature. These results enable each location and year to be considered an independent environment according to Romagosa and Fox (1993) in agreement with the results reported by Vaid et al. (1985) and Escribano et al. (1994). Nienhuis and Singh (1986) and Sills and Nienhuis (1993) have reported environmental effects in the expression of quantitative traits in dry and snap beans. Interactions between genotype and environment highlight the different response of the bean landraces to the environmental conditions. Mean values of pod weight and length indicate a trend to large pod in the

landraces, which implies a chance for snap bean breeding. Additionally the mean of pod curvature indicates that the general trend is for a straight pod, more valuable for breeding. Wide ranges of variation in all the characters except for pod curvature emphasize strongly the possibilities for selection inside this germplasm to obtain new inbred lines.

Values of constancy (C) displayed in Table 3 are in general low. Constancy values over 1.0 were found for the ratio width/thickness that indicated the pod shape (round or flat). Therefore, the pod shape is expressed clearly enough under different environmental conditions, with relevance for the selection within landraces of new inbred lines with specific shape.

The differences in the expression of the quantitative traits across the six environments could be explained over the basis of climatic factors. They could be responsible for the delayed flowering time and the pod maturity in Vitoria 1993 and Vitoria 1994 causing also long and heavy pods, as displayed in Table 4. On the other hand the period of flowering and the pod maturity in the other locations, Pontevedra and Lalín were short resulting in small to medium pods.

A relevant finding of this work arises from the combined analysis of variance and the principal component analysis. Environmental effects could not be assigned to different years (climatic factor) or locations (climatic and edaphic effect). The combined effect year by location means that each component one year-one location is in fact an independent factor (Romagosa and Fox 1993) affecting the horticultural value of the pod in snap bean.

Minimal data about snap bean production and recommendations for each area of production in Spain is available (Puerta-Romero 1961; Gascó 1971). According to the results of our research one must make a decision concerning the most adequate performance for snap bean use of the germplasm studied regarding pod traits and earliness. In the humid conditions of the North of Spain and Portugal earliness would permit hand-harvesting before autumn rains that could be regarded as a clear advantage for farmers. Concerning traits affecting pod shape, environmental effects appears to be not relevant.

The results of this work assess about the need to evaluate the new or improved snap bean varieties in different locations and years before making reliable recommendations to growers, as a consequence of the results displayed in Figure 1. A good performance of a landrace in a specific environment could be based upon earliness and large pods. Landraces showing this phenotype are distributed in the marked (by arrows) quarters of the six plots by environments. It means that Vitoria 1993 was less favourable for bean growing than the same location in 1994. Lalín had poor results in the performance of the landraces in the two years in spite of

small differences between years. Pontevedra seems to be the best environment for growing bean, but some of the best landraces are not repeated there from one year to another. It could be pointed out the presence of four landraces with good performances across five environments, being Lalín 1994 the less favourable for them.

As a conclusion, there is a considerable amount of variation available for the genetic improvement of snap bean cultivars and the enlargement of their genetic basis. Since some specific traits affecting pod quality are depending upon the effect of a few genes, a consistent genetic background coming from adapted landraces would support the breeding efforts to enlarge the genetic basis of the current snap bean cultivars. Therefore some adapted dry and double-use bean landraces from Spain and Portugal could contribute with valuable genetic background to snap bean breeding. Selection inside these landraces would be possible since variation intra-landrace often exists as reported in different sources of germplasm by Escribano et al. (1994), González et al. (1995), Traka-Mavrona (2000), and Rodiño et al. (2001). Varietal mixtures are sometimes seen in gardens and in markets in many regions and they could be separated into different lines according to seed colour and pattern. Specific pod traits that could give added value to inbred lines derived from adapted landraces merit further research and they could be introgressed to other bean cultivars by means of conventional breeding based on hybridization and backcrosses.

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Growth habit ¹								
Ι	II	III	IV					
Number of landraces (%)								
47 (38.9)	12 (9.9)	9 (7.4)	53 (43.8)					
	Pod color							
Green	Green-purple	Yellow						
Nu	Number of landraces (%)							
105 (86.8) 10 (8.2) 6 (5.0)								
	Fiber							
Low	ow Medium High							
Nu	Number of landraces (%)							
39 (32.2)	28 (23.2)	54 (44.6)						

Table 1. Characterization of three qualitative traits in the 121 bean landraces studied.

¹ I - determinate, II - indeterminate, upright, III- indeterminate, prostrate, IV - indeterminate, climbing

		Period of flowering	Pod maturity		Pod weight	Pod ler	ngth	Pod	Pod
		(days)	(days)		(g)	(mm)		width/thickness	curvature
Source of variation	df ¹				Mean squares				
Years (Y)	1	825.77	58502.1		44494.9	223061.1		0.1476	0.0540
Locations (L)	2	1781.91	213708.0		36517.9	29651.3		0.0494	0.0339
YxL	2	13710.02 **	26774.9	**	8765.7 **	43589.4	**	3.5404 **	0.0110
Rep YxL	6	206.06 **	154.2	**	450.9 **	937.8	**	0.2135 **	0.0110
Landraces (P)	120	176.72 **	362.8	**	767.9 **	5082.5	**	0.8217 **	0.0528 **
PxY	120	90.57 66.3		88.6	265.3		0.0604	0.010	01
PxL	240	104.27 72.9		97.5	* 237.4		0.0769	* 0.007	78
PxYxL	240	102.64 **	60.9	**	72.7 **	216.4	**	0.0594 *	0.0089
Error	720	39.94	29.0		49.5	147.1		0.0466	0.0081

Table 2. Analysis of variance mean, standard error (SE), range of variation and coefficient of variation (CV) of the quantitative traits evaluated in the 121 bean landraces studied.

Mean	28.2 80.5	7.07	122.1	2.14	ļ	1.049
SE	1.85	1.57	0.40	3.54	0.062	0.0266
Range of variation	18.4 - 38.0	70.1 - 101.3 3.7	2 - 13.34	75.7 - 193.9	1.13 - 3.05	0.854 - 1.318
CV (%)	22.5	6.7 3.92	9.9	10.1	l	8.7

¹ df: degrees of freedom

Components of	Period of	Pod	Pod	Pod	Pod width/	Pod
variance ¹	flowering	maturity	weight	length	thickness	curvature
	(days)	(days)	(g)	(mm)		
$\sigma^2 Y$	-17.730	46.38	52.21	262.3	-0.0050	0.0001
σ^2 L	-24.648	409.92	60.81	-30.6	-0.0078	0.0001
$\sigma^2{}_{\text{YxL}}$	55.542	116.62	36.37	186.8	0.0147	-0.0000
$\sigma^2_{R(YL)}$	1.373	1.10	3.52	6.9	0.0015	0.0000
σ^2_{P}	7.043	23.71	54.55	399.7	0.0620	0.0037
σ^2_{PxY}	-2.012	0.89	2.64	8.1	0.0002	0.0002
σ^2_{PxL}	0.408	3.04	6.19	5.2	0.0044	-0.0003
σ^2_{PxYxL}	31.353	15.99	11.59	34.7	0.0064	0.0004
σ^2	39.935	28.98	49.54	147.1	0.0466	0.0081
Ε(σ ² _P)	2.409	4.16	8.62	56.5	0.0092	0.0006
Variances ratio	2					
С	0.08	0.04	0.25	0.64	1.02	0.43
E(C)	0.03	0.01	0.04	0.09	0.15	0.07

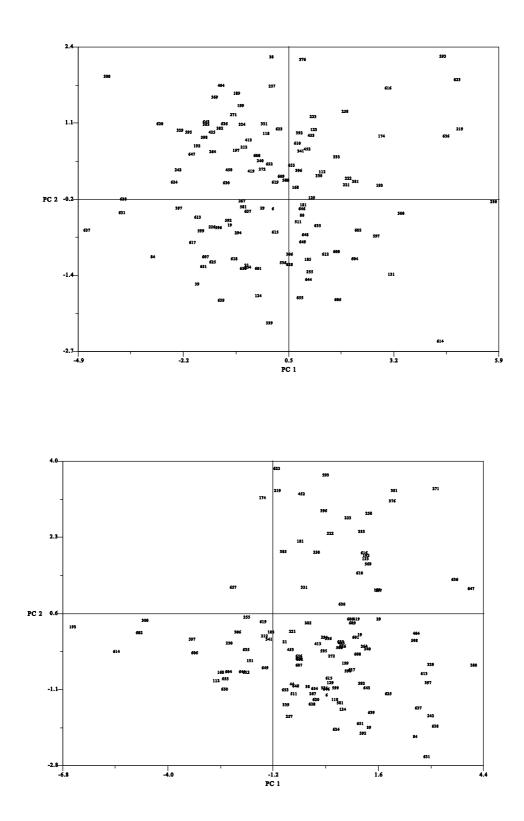
Table 3. Components and ratio of variances of the quantitative traits evaluated in the 121 bean landraces studied.

¹ σ_{Y}^{2} : variance of years; σ_{L}^{2} : variance of locations; σ_{YxL}^{2} : variance of interaction years by locations; $\sigma_{R(YL)}^{2}$: variance of replications in years and locations; σ_{P}^{2} : variance of landraces; σ_{PxY}^{2} : variance of the interaction landraces by years; σ_{PxL}^{2} : variance of the interaction landraces by locations; σ_{PxYxL}^{2} : variance of the interaction landraces by locations; σ_{PxYxL}^{2} : variance of the interaction landraces by locations; σ_{PxYxL}^{2} : variance of the interaction landraces by years and locations; σ_{PxYxL}^{2} : error variance. $E(\sigma_{P}^{2})$: error of the landraces variance. ² C (constancy) = $\sigma_{P}^{2} / (\sigma_{Y}^{2} + \sigma_{L}^{2} + \sigma_{YxL}^{2} + \sigma_{R(YL)}^{2} + \sigma_{PxY}^{2} + \sigma_{PxY}^{2} + \sigma_{PxYxL}^{2} + \sigma_$

Traits	Environments						
	Lalín	Lalín	Pontevedr	Pontevedr	Vitoria	Vitoria	
	1993	1994	a 1993	a 1994	1993	1994	
Period of	26.8	34.0	31.5	21.5	25.1	32.3	
flowering (days)							
Pod maturity	70.2	73.1	63.0	69.2	90.4	120.4	
(days)							
Pod weight	4.13	7.86	4.84	7.22	8.80	9.24	
(g)							
Pod length	92.6	137.5	111.4	130.7	127.0	135.2	
(mm)							
Pod width/	2.16	2.16	2.08	2.19	2.25	2.02	
thickness							

Table 4. Average across the six environments of five quantitative traits evaluated in the 121 bean landraces studied.

Figure 1. Ordination of the 121 landraces along the axis representing the first (PC 1), second (PC 2) and third (PC 3) principal components (numbers plus PHA- represent the code of the landraces) (the arrow indicates the quarter that includes the landraces with the best performance).



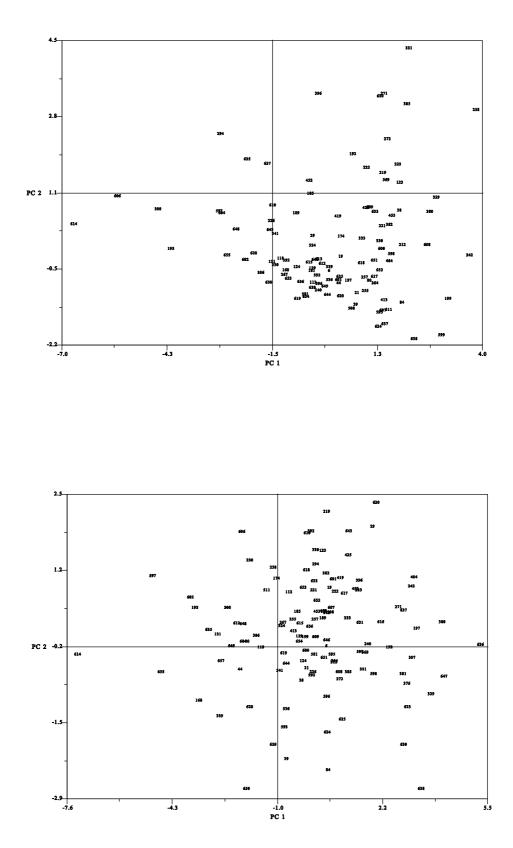


Figure 1. Continuation

