

Genetic variability among common black bean (*Phaseolus vulgaris* L.) accessions in southern Brazil

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Abstract

To obtain superior genotypes, the use of natural genetic variability is essential, aiming to select parents that will make future crossings blocks. Therefore, the aim of this study was to determine the genetic variability and dissimilarity using multivariate analyzes in common black bean accessions native to southern Brazil. The experiment was conducted in the 2014/2015 in Frederico Westphalen – RS. The experimental design was augmented blocks (RCBD) with four repetitions (totally 149 treatments), in which 147 black common bean accessions were evaluated. Two commercial cultivars were used as witnesses: BRS Esplendor and BRS Supremo. The phenotypic traits such as days to flowering, plant height at flowering, days to maturity, plant height at maturity, insertion of the first pod, number of pods per plant, number of seeds per pod, mass of seeds per pod, number of branches, mass of seeds per plant, seed length, seed width, seed flattening, seed brightness, presence of halo, color of the halo were measured. The distributions of phenotypic frequencies indicate genetic variability among the 149 genotypes of common black bean. The Tocher optimization method presents the formation of eight groups of genotypes. The dendrogram formed by the standardized Euclidean distance was efficient in the stratification of the accessions for their genetic distance. The relative contribution evaluated by Singh method shows that the characters days to flowering and seed brightness are those that best discriminate the genotypes. The multivariate techniques of Tocher optimization and the standardized Euclidean distance show similar responses, proving to be viable tools for the choice of parents in a breeding program.

Keywords: genetic breeding; multivariate analyses; landraces; *Phaseolus vulgaris* L.

Abbreviations: DTF_days to flowering; PHF_plant height at flowering; DTM_days to maturity; PHM_plant height at maturity; IFP_insertion of the first pod; NPP_number of pods per plant; NSP_number of seeds per pod; MSP_mass of seeds per pod; NB_number of branches; MSPP_mass of seeds per plant; SL_seed length; SW_seed width; SF_seed flattening; BRI_seed brightness; HAL_presence of halo; CRH_color of the halo; cm_centimeters; UPGMA_grouping of unweighed pairs based on the arithmetic mean; MC_main components; CFA_subtropical; RCBD_experimental design was of blocks augmented; m_meters; m²_square meters; kg ha⁻¹_kilograms per hectare; V4_vegetative stage 4.

Introduction

The common black bean (*Phaseolus vulgaris* L.) is one of the major legumes intended for human consumption. It is grown in much of the Brazilian territory (Elias et al., 2007). The bean nutritional capacity is due to genotype, environment and genotype × environment interaction. The main components of nutrition are proteins, starch, sugar and vitamins (Flores et al., 2009). The genetic breeding of the species seeks genotypes that do not suffer from the environmental factors and interaction of genotype × environment. At the same time, they must have high production levels, cooking quality and high nutritional content (Carbonell et al., 2003).

The genetic breeding program of black bean has defined an ideotype with 35 days to start flowering (Silva et al., 2007), plants with stature higher than 50 centimeters (Horn et al., 2010), physiological maturity of up to 90 days (Elias et al., 2007), greater height of insertion of the first pod (Rocha et al., 2014), high number of pods per plant and seeds per pod (Kurek et al., 2001), reflecting high productivity (Ramos

Junior et al., 2005). Therefore, to obtain superior genotypes, it is essential to use the natural genetic variability to select efficient parents that will make future crossings programs. Hybridization between the genetically distant genotypes can enhance the genetic variability of the segregating populations and increase the chance of obtaining transgressive genotypes (Ramalho et al., 2012). Molecular techniques are used as effective tools to identify genes and alleles responsible for the traits of interest, selection of higher yielding genotypes, pyramiding gene, preparation of genetic maps and to identify the genetic variability of common black beans (Marin et al., 2005). Thus, understanding the genetic distance between the common black bean genotypes is crucial, where contrasting parents with additional and favorable traits are sought.

Among the multivariate analyses, the Tocher optimization method aims at subdividing the genotypes into groups, with the lowest genetic dissimilarity within and the highest dissimilarity among groups. The standardized Euclidean distance seeks through a set of observations to discriminate

genotypes by forming groups with close phenotypes (Cruz et al., 2012). The methodology proposed by Singh (1981) can reveal the relative contribution of each character for discrimination of genotypes. The principal component analysis allows generating independent information and shows how many variables are needed to explain the genetic variation among genotypes (Cruz et al., 2012).

The aim of this study was to determine the genetic variability and dissimilarity by multivariate analyzes in common black bean accessions native to southern Brazil.

Results and Discussion

Characterization of traits

The descriptive analysis revealed the formation of different classes among the 149 genotypes for the traits days to flowering (DTF), plant height at flowering (PHF), days to maturity (DTM), plant height at maturity (PHM), insertion of the first pod (IFP), number of pods per plant (NPP), number of seeds per pod (NSP), mass of seeds per pod (MSP), number of branches (NB), mass of seeds per plant (MSPP), seed length (SL), seed width (SW), seed flattening (SF), seed brightness (BRI), presence of halo (HAL), and color of the halo (CRH). The range of coefficients of variation of characters was 8.13% to 35.71%, which shows adequate performance of the experiment.

Performance of traits on the frequency of analysis

The character days to flowering (DTF) revealed the formation of seven phenotypic classes with an amplitude of 25 to 55 days (Fig. 1). Classes of 40 and 45 DTF encompassed 74.5% of the genotypes. Genetic breeding programs aim to get early genotypes, with up to 35 DTF, requiring less water and nutrient resources. Given this, the selection of this character appears as the most viable way to define the common black bean cycle, controlled by a few genes with high heritability (Silva et al., 2007). Therefore, the selection for earliness can be proceeded on the phenotypic classes of 25, 30 and 35 DTF, consist of 23.75% of the studied genotypes.

The plant height at flowering (PHF) presented seven phenotypic classes, ranging from 15 to 85 cm (Fig. 1), wherein the classes of 15 and 25 cm consist of 70% of the genotypes. The agronomic ideotype favorable to the mechanical harvest must have plants with stature higher than 50.0 cm and erect (Horn et al., 2000). The character days to maturity (DTM) expressed eight phenotypic classes and amplitude from 72 to 114 DTM (Fig. 1). The class 102 DTM represents 48.3% of the genotypes. Research carried out with 45 common black bean genotypes determined the average length of 87.4 days (Elias et al., 2007). Therefore, the selection for earlier mature genotypes can occur by means of the phenotypic classes 72, 78 and 84 DTM.

Plant height at maturity (PHM) presented eight phenotypic classes ranging from 15 to 120 cm (Fig.1), wherein the class of 45 cm encompassed 36.9% of the genotypes. Studies to identify the best space arrangements for different bean genotypes showed final height of 54.1 cm, concluding that the increase of this character have a negative impact on productivity (Morais et al., 2001). Thus, agronomically favorable genotypes can be obtained from the phenotypic classes of 45 to 60 cm.

For the character insertion of the first pod (IFP), ten phenotypic classes with range of 3-30 cm were found (Fig.1).

The class of 15 cm stood out, with 22.8% of genotypes. Diallel analysis of bean genotypes advocate for genotypes with more IFP, and define that the genetic gain for this character was determined by the efficient choice of parents (Rocha et al., 2014). The selection to increment this character can occur in the classes 15, 18, 21, 24, 27 and 30 cm.

The number of pods per plant (NPP) showed nine phenotypic classes, ranging 2 to 34 pods (Fig. 1). The class with six pods per plant accounted for 34.2% of genotypes. Multivariate analysis for 32 black bean genotypes indicated that the height and duration of the common black bean cycle are essential to increase the number of pods and the mass of seeds per plant (Coimbra et al., 2000). Increased productivity can be obtained by the phenotypic class with 34 pods per plant.

Regarding the number of seeds per pod (NSP), nine phenotypic classes were observed, with pods ranging from 1.2 to 6.0 seeds (Fig. 1). The class 4.2 seeds per pod included the largest fraction of the genotypes. The selection can be directed to the class with six seeds per pod. The mass of seeds per pod (MSP) revealed eight phenotypic classes ranging from 0.38 to 2.13 grams (Fig.1). However, 32.2% of the genotypes were indexed in the class with 0.63 grams. A less contribution of NSP is attributed to the characteristics that contribute to more productive genotypes; however, more attention is given to the NPP (Kurek et al., 2001).

The number of branches (NB) presents seven phenotypic classes, with a range of 0.4 to 6.0 branches per plant (Fig. 2), and 32.2% of the genotypes reside in the lowest class. For the mass of seeds per plant (MSPP), nine phenotypic classes are formed, ranging from 3.0 to 27.0 grams of seed per plant (Fig. 2). The largest proportion of genotypes was located in the class with 6.0 grams, corresponding to 28.8% of genotypes. So, the selection of superior genotypes must be grounded in the characters mass of seeds per plant and number of seeds per pod. These are crucial to the productivity of the common black bean (Ramos Junior et al., 2005). Hence, obtaining more productive genotypes can be achieved in the phenotypic classes with greater magnitude of this character.

Seed length (SL) revealed eight phenotypic classes with a range of 6.0 to 13.0 millimeters. Of these, 32.2% of the genotypes were indexed in the class with 10.0 mm (Fig. 2). Seed width (SW) shows seven phenotypic classes, comprising from 3.9 to 8.1 mm, in which 38.9% of the genotypes stood in the 6.3 mm class (Fig. 2). Silva (2005) classifies the shape of seeds by the ratio between length and width. The greater proportion of genotypes expressed ratio of 1.53 and the seed shape was assigned as elliptical.

As for the the visual characteristics determined by Silva (2005), we found that for the seed flattening (SF), 83.0% of the genotypes were classified as moderately filled seeds (class 2.0), 15% as filled seeds (class 2.8) and 2.0% as flattened seeds (class 5.2) (Fig. 2). Regarding the brightness of the seed (BRI), 90% of the genotypes showed seeds with average brightness (class 2.03) and 10.0% had opaque seeds (class 0.83) (Fig. 2). The character presence of halo (HAL) shows that 93.0% of the genotypes do not have halo (class 1.02) (Fig. 2). For the color of halo (CRH), 99.0% of the genotypes express halo color similar to the rest of the seed (class 1.02).

Performance of traits for grouping optimization Tocher

The Tocher optimization method was performed for the 147 common black bean accessions and for two commercial

Table 1. Additional information and origin of the common black bean access.

Access identification	Genotypes	Population source	City of origin	Geographic coordinates
1	LMGPP1	1	Campos Borges, RS, Brazil	
2	LMGPP2	5	Campos Borges, RS, Brazil	
3	LMGPP3	6	Campos Borges, RS, Brazil	
4	LMGPP4	6	Campos Borges, RS, Brazil	
5	LMGPP5	8	Campos Borges, RS, Brazil	28° 52' 31"S and 53° 00' 55"W
6	LMGPP6	8	Campos Borges, RS, Brazil	
7	LMGPP7	8	Campos Borges, RS, Brazil	
8	LMGPP8	8	Campos Borges, RS, Brazil	
9	LMGPP9	9	Campos Borges, RS, Brazil	
10	LMGPP10	9	Campos Borges, RS, Brazil	
11	LMGPP11	10	Palmeira das Missões, RS, Brazil	
12	LMGPP12	12	Palmeira das Missões, RS, Brazil	
13	LMGPP13	12	Palmeira das Missões, RS, Brazil	
14	LMGPP14	13	Palmeira das Missões, RS, Brazil	
15	LMGPP15	13	Palmeira das Missões, RS, Brazil	
16	LMGPP16	13	Palmeira das Missões, RS, Brazil	
17	LMGPP17	13	Palmeira das Missões, RS, Brazil	
18	LMGPP18	13	Palmeira das Missões, RS, Brazil	
19	LMGPP19	13	Palmeira das Missões, RS, Brazil	
20	LMGPP20	13	Palmeira das Missões, RS, Brazil	27° 53' 19"S and 53° 18' 19" W
21	LMGPP21	13	Palmeira das Missões, RS, Brazil	
22	LMGPP22	16	Palmeira das Missões, RS, Brazil	
23	LMGPP23	16	Palmeira das Missões, RS, Brazil	
24	LMGPP24	16	Palmeira das Missões, RS, Brazil	
25	LMGPP25	16	Palmeira das Missões, RS, Brazil	
26	LMGPP26	16	Palmeira das Missões, RS, Brazil	
27	LMGPP27	16	Palmeira das Missões, RS, Brazil	
28	LMGPP28	16	Palmeira das Missões, RS, Brazil	
29	LMGPP29	20	Palmeira das Missões, RS, Brazil	
30	LMGPP30	20	Palmeira das Missões, RS, Brazil	

Access identification	Genotypes	Population source	City of origin	Geographic coordinates
31	LMGPP31	21	Santa Rosa, RS, Brazil	
32	LMGPP32	24	Santa Rosa, RS, Brazil	
33	LMGPP33	24	Santa Rosa, RS, Brazil	
34	LMGPP34	25	Santa Rosa, RS, Brazil	
35	LMGPP35	25	Santa Rosa, RS, Brazil	
36	LMGPP36	38	Santa Rosa, RS, Brazil	
37	LMGPP37	39	Santa Rosa, RS, Brazil	
38	LMGPP38	39	Santa Rosa, RS, Brazil	
39	LMGPP39	39	Santa Rosa, RS, Brazil	
40	LMGPP40	40	Santa Rosa, RS, Brazil	
41	LMGPP41	40	Santa Rosa, RS, Brazil	
42	LMGPP42	40	Santa Rosa, RS, Brazil	
43	LMGPP43	40	Santa Rosa, RS, Brazil	
44	LMGPP44	41	Santa Rosa, RS, Brazil	27° 52' 16" S and 54° 28' 55" W
45	LMGPP45	41	Santa Rosa, RS, Brazil	
46	LMGPP46	41	Santa Rosa, RS, Brazil	
47	LMGPP47	41	Santa Rosa, RS, Brazil	
48	LMGPP48	41	Santa Rosa, RS, Brazil	
49	LMGPP49	41	Santa Rosa, RS, Brazil	
50	LMGPP50	41	Santa Rosa, RS, Brazil	
51	LMGPP51	41	Santa Rosa, RS, Brazil	
52	LMGPP52	47	Santa Rosa, RS, Brazil	
53	LMGPP53	49	Santa Rosa, RS, Brazil	
54	LMGPP54	49	Santa Rosa, RS, Brazil	
55	LMGPP55	49	Santa Rosa, RS, Brazil	
56	LMGPP56	49	Santa Rosa, RS, Brazil	
57	LMGPP57	49	Santa Rosa, RS, Brazil	
58	LMGPP58	50	Pejuçara, RS, Brazil	
59	LMGPP59	50	Pejuçara, RS, Brazil	28° 25' 24" S and 53° 39' 21" W
60	LMGPP60	52	Pejuçara, RS, Brazil	

Continue.

Access identification	Genotypes	Population source	City of origin	Geographic coordinates
61	LMGPP61	52	Pejuçara, RS, Brazil	28° 25' 24" S and 53° 39' 21"W
62	LMGPP62	55	Pejuçara, RS, Brazil	
63	LMGPP63	55	Pejuçara, RS, Brazil	
64	LMGPP64	55	Pejuçara, RS, Brazil	
65	LMGPP65	58	Pejuçara, RS, Brazil	
66	LMGPP66	59	Pejuçara, RS, Brazil	
67	LMGPP67	64	Pejuçara, RS, Brazil	
68	LMGPP68	64	Pejuçara, RS, Brazil	
69	LMGPP69	64	Pejuçara, RS, Brazil	
70	LMGPP70	64	Pejuçara, RS, Brazil	
71	LMGPP71	64	Pejuçara, RS, Brazil	
72	LMGPP72	64	Pejuçara, RS, Brazil	
73	LMGPP73	64	Pejuçara, RS, Brazil	
74	LMGPP74	64	Pejuçara, RS, Brazil	
75	LMGPP75	64	Pejuçara, RS, Brazil	
76	LMGPP76	64	Pejuçara, RS, Brazil	
77	LMGPP77	64	Pejuçara, RS, Brazil	
78	LMGPP78	64	Pejuçara, RS, Brazil	
79	LMGPP79	64	Pejuçara, RS, Brazil	
80	LMGPP80	64	Pejuçara, RS, Brazil	
81	LMGPP81	64	Pejuçara, RS, Brazil	
82	LMGPP82	64	Pejuçara, RS, Brazil	
83	LMGPP83	64	Pejuçara, RS, Brazil	
84	LMGPP84	64	Pejuçara, RS, Brazil	
85	LMGPP85	64	Pejuçara, RS, Brazil	
86	LMGPP86	64	Pejuçara, RS, Brazil	
87	LMGPP87	64	Pejuçara, RS, Brazil	
88	LMGPP88	64	Pejuçara, RS, Brazil	
89	LMGPP89	64	Pejuçara, RS, Brazil	
90	LMGPP90	64	Pejuçara, RS, Brazil	

Access identification	Genotypes	Population source	City of origin	Geographic coordinates
61	LMGPP61	52	Pejuçara, RS, Brazil	28° 25' 24" S and 53° 39' 21"W
62	LMGPP62	55	Pejuçara, RS, Brazil	
63	LMGPP63	55	Pejuçara, RS, Brazil	
64	LMGPP64	55	Pejuçara, RS, Brazil	
65	LMGPP65	58	Pejuçara, RS, Brazil	
66	LMGPP66	59	Pejuçara, RS, Brazil	
67	LMGPP67	64	Pejuçara, RS, Brazil	
68	LMGPP68	64	Pejuçara, RS, Brazil	
69	LMGPP69	64	Pejuçara, RS, Brazil	
70	LMGPP70	64	Pejuçara, RS, Brazil	
71	LMGPP71	64	Pejuçara, RS, Brazil	
72	LMGPP72	64	Pejuçara, RS, Brazil	
73	LMGPP73	64	Pejuçara, RS, Brazil	
74	LMGPP74	64	Pejuçara, RS, Brazil	
75	LMGPP75	64	Pejuçara, RS, Brazil	
76	LMGPP76	64	Pejuçara, RS, Brazil	
77	LMGPP77	64	Pejuçara, RS, Brazil	
78	LMGPP78	64	Pejuçara, RS, Brazil	
79	LMGPP79	64	Pejuçara, RS, Brazil	
80	LMGPP80	64	Pejuçara, RS, Brazil	
81	LMGPP81	64	Pejuçara, RS, Brazil	
82	LMGPP82	64	Pejuçara, RS, Brazil	
83	LMGPP83	64	Pejuçara, RS, Brazil	
84	LMGPP84	64	Pejuçara, RS, Brazil	
85	LMGPP85	64	Pejuçara, RS, Brazil	
86	LMGPP86	64	Pejuçara, RS, Brazil	
87	LMGPP87	64	Pejuçara, RS, Brazil	
88	LMGPP88	64	Pejuçara, RS, Brazil	
89	LMGPP89	64	Pejuçara, RS, Brazil	
90	LMGPP90	64	Pejuçara, RS, Brazil	

Access identification	Genotypes	Population source	City of origin	Geographic coordinates
91	LMGPP91	64	Pejuçara, RS, Brazil	
92	LMGPP92	67	Braga, RS, Brazil	
93	LMGPP93	68	Braga, RS, Brazil	
94	LMGPP94	68	Braga, RS, Brazil	
95	LMGPP95	69	Braga, RS, Brazil	
96	LMGPP96	69	Braga, RS, Brazil	
97	LMGPP97	69	Braga, RS, Brazil	
98	LMGPP98	69	Braga, RS, Brazil	
99	LMGPP99	72	Braga, RS, Brazil	27° 37' 16" S and 53° 44' 17" W
100	LMGPP100	72	Braga, RS, Brazil	
101	LMGPP101	72	Braga, RS, Brazil	
102	LMGPP102	72	Braga, RS, Brazil	
103	LMGPP103	72	Braga, RS, Brazil	
104	LMGPP104	72	Braga, RS, Brazil	
105	LMGPP105	72	Braga, RS, Brazil	
106	LMGPP106	73	Cruz Alta, RS, Brazil	
107	LMGPP107	73	Cruz Alta, RS, Brazil	
108	LMGPP108	73	Cruz Alta, RS, Brazil	
109	LMGPP109	73	Cruz Alta, RS, Brazil	
110	LMGPP110	73	Cruz Alta, RS, Brazil	
111	LMGPP111	73	Cruz Alta, RS, Brazil	
112	LMGPP112	73	Cruz Alta, RS, Brazil	
113	LMGPP113	74	Cruz Alta, RS, Brazil	28° 38' 22" S and 53° 36' 22" W
114	LMGPP114	74	Cruz Alta, RS, Brazil	
115	LMGPP115	74	Cruz Alta, RS, Brazil	
116	LMGPP116	74	Cruz Alta, RS, Brazil	
117	LMGPP117	74	Cruz Alta, RS, Brazil	
118	LMGPP118	74	Cruz Alta, RS, Brazil	
119	LMGPP119	74	Cruz Alta, RS, Brazil	
120	LMGPP120	74	Cruz Alta, RS, Brazil	

Access Identification	Genotypes	Population source	City of origin	Geographic coordinates
121	LMGPP121	74	Cruz Alta, RS, Brazil	
122	LMGPP122	74	Cruz Alta, RS, Brazil	
123	LMGPP123	74	Cruz Alta, RS, Brazil	
124	LMGPP124	75	Cruz Alta, RS, Brazil	
125	LMGPP125	75	Cruz Alta, RS, Brazil	
126	LMGPP126	75	Cruz Alta, RS, Brazil	28° 38' 22" S and 53° 36' 22" W
127	LMGPP127	75	Cruz Alta, RS, Brazil	
128	LMGPP128	75	Cruz Alta, RS, Brazil	
129	LMGPP129	75	Cruz Alta, RS, Brazil	
130	LMGPP130	75	Cruz Alta, RS, Brazil	
131	LMGPP131	75	Cruz Alta, RS, Brazil	
132	LMGPP132	76	Fortaleza dos Valos, RS, Brazil	
133	LMGPP133	76	Fortaleza dos Valos, RS, Brazil	
134	LMGPP134	76	Fortaleza dos Valos, RS, Brazil	
135	LMGPP135	76	Fortaleza dos Valos, RS, Brazil	
136	LMGPP136	76	Fortaleza dos Valos, RS, Brazil	
137	LMGPP137	76	Fortaleza dos Valos, RS, Brazil	28° 46' 60" S and 53° 13' 24" W
138	LMGPP138	76	Fortaleza dos Valos, RS, Brazil	
139	LMGPP139	76	Fortaleza dos Valos, RS, Brazil	
140	LMGPP140	76	Fortaleza dos Valos, RS, Brazil	
141	LMGPP141	76	Fortaleza dos Valos, RS, Brazil	
142	LMGPP142	8	Campos Borges, RS, Brazil	
143	LMGPP143	8	Campos Borges, RS, Brazil	
144	LMGPP144	8	Campos Borges, RS, Brazil	
145	LMGPP145	8	Campos Borges, RS, Brazil	28° 52' 31" S and 53° 00' 55" W
146	LMGPP146	8	Campos Borges, RS, Brazil	
147	LMGPP147	8	Campos Borges, RS, Brazil	
148	BRS Esplendor	Commercial cultivars witnesses	.	.
149	BRS Supremo	Commercial cultivars witnesses	.	.

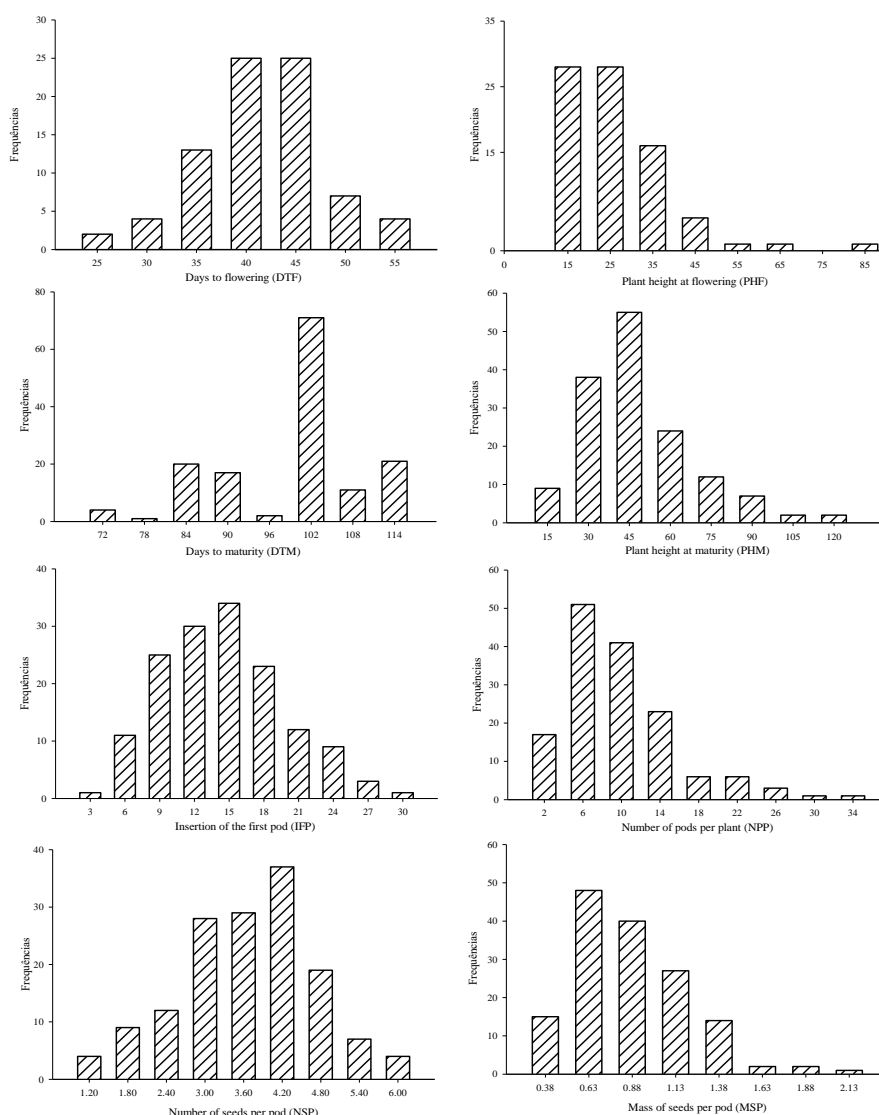


Fig 1. Distribution of frequencies of the phenotypic classes for, graph (A) days to flowering (DTF); graph (B) plant height at flowering (PHF); graph (C) days to maturity (DTM); graph (D) plant height at maturity (PHM); graph (E) insertion of the first pod (IFP); graph (F) number of pods per plant (NPP); graph (G) number of seeds per pod (NSP); graph (H) mass of seeds per pod (MSP), measured in the 2014/2015 crop.

cultivars BRS Esplendor and BRS Supremo, based on the standardized Euclidean distance (Table 2). This method has the purpose of subdividing the genotypes into groups, using the criterion of maintaining the lowest genetic dissimilarity within the group and the greatest dissimilarity between groups. In this way, genotypes have the lowest genetic distance from the first group (Cruz et al., 2012).

The genotypes were subdivided into eight groups. The group I consisting of 120 black bean accessions and the commercial cultivars BRS Esplendor and BRS Supremo, where the greatest genetic distances for group I were observed between the accessions 43 and 105. The group II consisted of 13 genotypes; group III, six genotypes; group IV three genotypes; group V two genotypes; and the groups VI, VII and VIII were consisted of only one genotype each. Research about 45 black bean genotypes showed great contribution of Tocher grouping to differentiate genotypes and direct future crossings between the groups more genetically distant (Elias et al., 2007).

Regarding the genetic distance between the groups (Table

3), higher means were observed between the groups V and VIII (2.27) and VI and VIII (2.21). Therefore, it is possible to increase the genetic variability of the breeding program, using the genotypes from more genetically distant groups. One alternative is to direct the crossings between the individuals of group I with group VIII (1.84), as in the first group there are accessions with agronomic ideotypes closer to the ideal phenotype of the commercial cultivars BRS Esplendor and BRS Supremo.

Performance of traits for standardized Euclidean distance

The standardized Euclidean distance is obtained through a collection of information measured by means of several characters evaluated in each individual (Cruz et al., 2012). The dendrogram shows the genetic dissimilarity between 147 accessions and the cultivars BRS Esplendor (148) and BRS Supremo (149). It was found that the accession 139 is characterized as the most dissimilar genotype, followed by

Table 2. Results for the grouping of 147 common black bean accessions and controls BRS Esplendor and BRS Supremo, by the Tocher optimization method, based on the standardized Euclidean distance, considering of 16 characteristics of agronomic importance measured in the 2014/2015 crop.

Groups	Common black bean genotypes
I	43, 46, 45, 41, 40, 47, 124, 108, 146, 144, 39, 129, 107, 141, 71, 15, 66, 133, 42, 69, 1, 12, 123, 38, 145, BRS Esplendor, 84, 77, 62, 31, 92, 28, 54, 20, 27, 74, 135, 91, 138, 118, 117, 106, 70, 16, 140, 121, 19, 127, 101, 32, 50, 44, 137, 7,
II	52, 58, 14, 37, 18, 119, 8, 34, 96, 128, 72, BRS Supremo, 57, 132, 82, 93, 13, 89, 116, 109, 5, 3, 30, 63, 49, 88, 130, 25, 48, 80, 120, 75, 125, 61, 76, 68, 65, 21, 126, 23, 131, 142, 78, 73, 22, 53, 67, 24, 6, 59, 83, 2, 10, 35, 60, 99, 90, 143, 134, 29, 9, 11, 95, 79, 122, 33, 100, 105
III	104, 115, 86, 97, 55, 113, 114, 26, 85, 103, 112, 147, 102
IV	17, 87, 4, 36, 111, 136
V	64, 81, 98
VI	94, 110
VII	51
VIII	56
	139

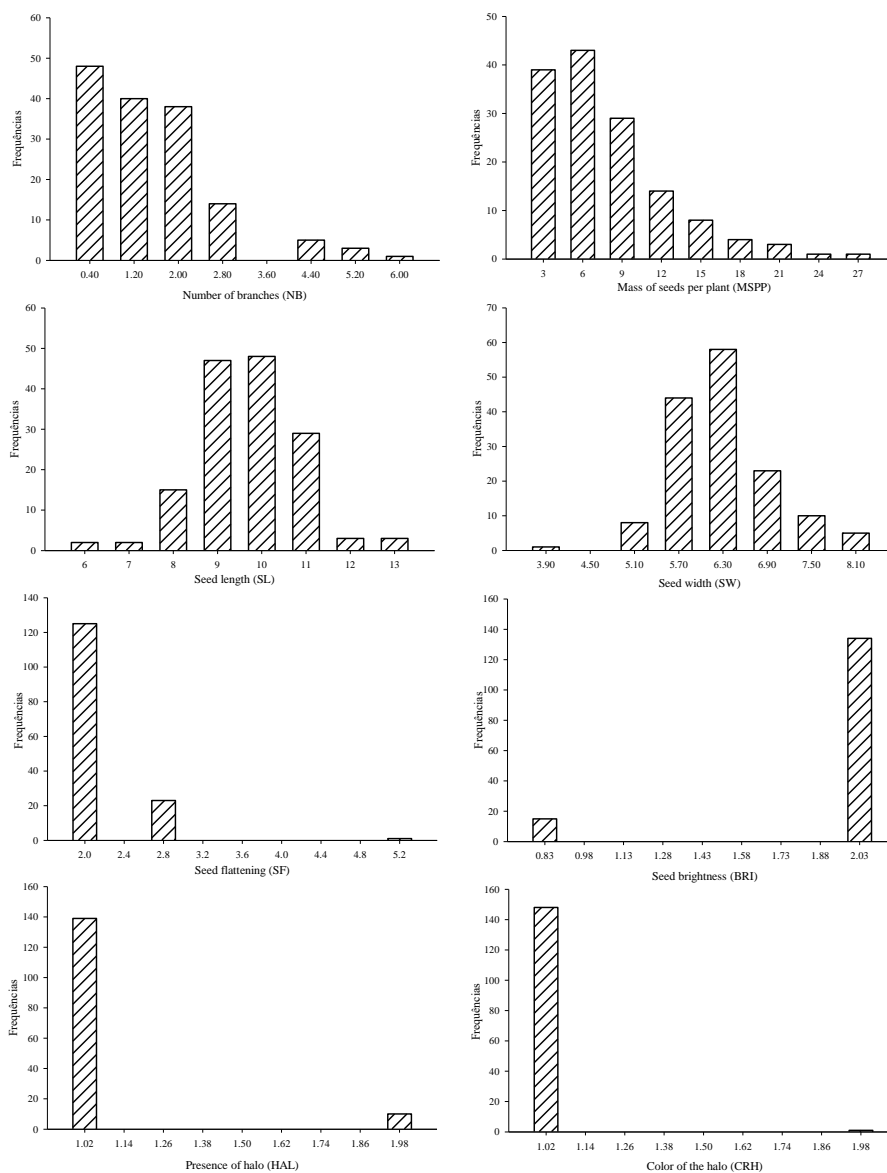


Fig 2. Distribution of frequencies of the phenotypic classes for, graph (A) number of branches (NB); graph (B) mass of seeds per plant (MSPP); graph (C) seed length (SL); graph (D) seed width (SW); graph (E) seed flattening (SF); graph (F) seed brightness (BRI); graph (G) presence of halo (HAL); graph (H) color of the halo (CRH), measured in the 2014/2015 crop.

Table 3. Results of the average distance between groups estimated by the Tocher optimization method, involving 147 common black bean accessions and controls BRS Esplendor and BRS Supremo, considering 16 characteristics of agronomic importance measured in the 2014/2015 crop.

Grupos	I	II	III	IV	V	VI	VII	VIII
I	0.80	1.35	1.23	1.38	1.27	1.33	1.29	1.84
II		0.93	1.49	1.56	1.65	1.74	1.66	1.56
III			0.67	1.58	1.52	1.63	1.60	2.08
IV				0.92	1.45	1.61	1.73	2.07
V					0.92	1.37	1.56	2.27
VI						-	1.51	2.21
VII							-	2.05
VIII								-

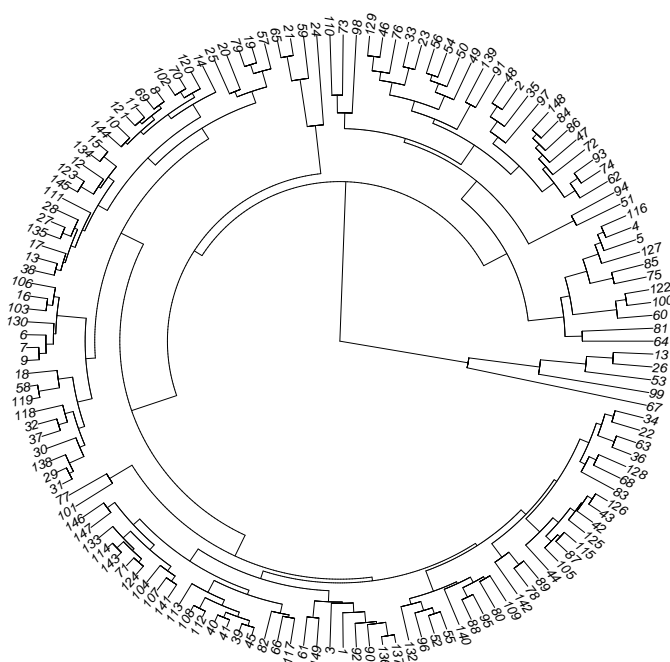


Fig3. Dendrogram with the genetic dissimilarity in 147 common black bean accessions and controls BRS Esplendor (148) and BRS Supremo (149), using the standardized Euclidean distance, obtained by the method of average connections (UPGMA), measured in the 2014/2015crop.

accessions 51 and 56 (Fig. 3). From these results, it is possible to identify that the grouping of unweighted pairs based on the arithmetic mean (UPGMA) shows similar results to those obtained by the Tocher optimization method, being possible to identify the formation of the eight groups, in which groups I, II, III IV, V, VI, VII and VIII are comprised of 122, 13, 6, 3, 2, 1, 1, and 1 genotype, respectively. Comparison of clustering methods for bean genotypes concluded that the responses obtained by the standardized Euclidean distance are similar to those obtained by the Tocher optimization method (Cargnelutti Filho et al., 2008).

Performance of traits for the relative contribution by Singh

The relative contribution of the characters in the discrimination of genotypes was performed by Singh method (1981), using 16 characters measured in 147 accessions and two check cultivars, BRS Esplendor and BRS Supremo (Table 4). Thus, the variables with the highest relative

contribution were days to flowering (DTF), with 21.93%, and seed brightness (BRI) with 13.31%. Studies in 57 black bean accessions concluded that the highest relative contributions to differentiate the genotypes are obtained by mass of hundred seeds, days to flowering and maturity, and seed size (Cabral et al., 2011).

Performance of traits for the principal components

The principal component analysis is characterized as a multivariate technique that allows, by means of a set of characters generating independent information, to explain and infer a response in the most informative way (Cruz et al., 2012). Given this, nine main components were needed to explain the genetic variation between the common black bean genotypes, where MC 1 explained (18.3%), MC 2 (12.8%), MC 3 (10.0%), MC 4 (8.5%), MC 5 (7.9%), MC 6 (6.8%), MC 7 (6.1%), MC 8 (5.9%) and MC 9 (5.2%). These components together made it possible to explain 81.9% of all genetic variation involved in the 149 genotypes studied.

Table 4. Results of the relative contribution of 147 common black bean accessions and controls BRS Esplendor and BRS Supremo, by the method of Singh (1981). Measured in the 2014/2015 crop.

Variable	S.j	Relative Contribution (%)
Days to flowering (DTF)	3303.72	21.93
Plant height at flowering (PHF)	958.34	6.36
Days to maturity (DTM)	343.51	2.28
Plant height at maturity (PHM)	701.24	4.65
Insertion of the first pod (IFP)	866.33	5.75
Number of pods per plant (NPP)	710.68	4.72
Number of seeds per pod (NSP)	947.64	6.29
Mass of seeds per pod (MSP)	688.61	4.57
Number of branches (NB)	1006.67	6.68
Mass of seeds per plant (MSPP)	730.60	4.85
Seed length (SL)	535.45	3.55
Seed width (SW)	430.59	2.86
Seed flattening (SF)	300.54	1.99
Seed brightness (BRI)	2005.00	13.31
Presence of halo (HAL)	1390.00	9.23
Color of the halo (CRH)	148.00	0.98

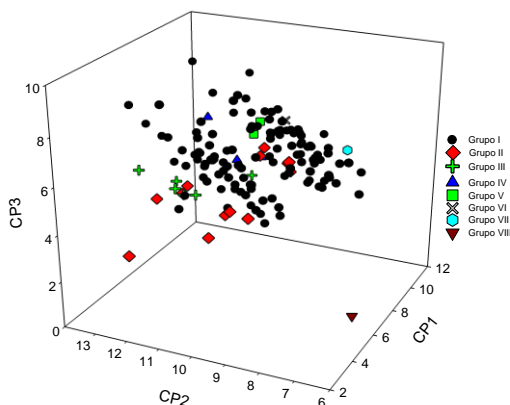


Fig 4. Results for the analysis of graphic dispersion of the scores in relation to the first three main components, MC1, MC2 and MC3, of the 147 common black bean accessions and controls BRS Esplendor and BRS Supremo, measured in the 2014/2015 crop.

Regarding the graphic dispersion of the accessions, only the three first main components (MC1, MC2, and MC3) were used to demonstrate the dispersion of genotypes (Fig 4). To better understand the distinction of genotypes, the Tocher optimization and classification was used, assigning distinct evidence to the eight formed groups. Based on the results obtained in each multivariate analysis, it was found that the response of the 147 accessions and the check cultivars (BRS Esplendor and BRS Supremo) are similar in Tocher optimization method, the standardized Euclidean distance and the principal component analysis. So, they were efficient to distinguish genotypes, being characterized as viable tools to assist in the selection of genotypes that will compose crossings blocks of a breeding program for the common black bean culture.

Materials and Methods

Plant materials

The experiment was conducted in the 2014/2015 crop season in Frederico Westphalen-RS, where the coordinates correspond to 27°39' 56" S latitude and 53°42' 94" W longitude, with an altitude of 490 meters. The soil is classified as a Typic Alumino ferric Red Latosol and the climate is characterized, by Köppen, as subtropical (*Cfa*) (Moreno, 1961).

Experimental design and experimental procedure

The experimental design was an augmented blocks (RCBD) (Federer, 1956) with 147 black common bean accessions, and two commercial cultivars witnesses (BRS Esplendor and BRS Supremo) with four repetitions, totally 149 treatments (each treatment as a one genotype). The accessions are from landraces of common black bean collected in different regions of southern Brazil, and available in the Laboratory of Genetic Breeding and Plant Production of the Federal University of Santa Maria *Campus* Frederico Westphalen, RS, Brazil (Table 1). The experimental units were composed of two lines spaced by 0.45 meters and two meters (m) long, totally 1.80 m². The population density employed for all accessions and cultivars of common black bean was of 10 seeds per linear meter, according to Cabral et al. (2011). The management was employed based on direct sowing, with fertilizer in the sowing line of 300 kg ha⁻¹ of the formulated N-P-K (10-20-20). A 90 kg ha⁻¹ of nitrogen was applied in the amide form. The control of weeds, insect pests and diseases were carried out, preventively.

Traits measured

The evaluated traits were: Days to flowering (DTF), measured by counting the number of days from seedling emergence until the issuance of the first flower bud. Plant

height at flowering (PHF), measured from the ground level to the last fully expanded leaflet at the beginning of the reproductive period. Days to maturity (DTM) was measured by counting the number of days from seedling emergence to harvest. Plant height at maturity (PHM) was measured from the ground level to the apex of the plant measured at harvest. Insertion of the first pod (IFP) was measured by the distance between the ground level and the insertion of the first pod viable on the main stem. Number of pods per plant (NPP) was determined by counting the total number of viable pods per plant. Number of seeds per pod (NSP) was obtained by the ratio between the total number of viable pods and the number of seeds per plant, results in units. Mass of seeds per pod (MSP) was obtained by the ratio between the total mass of seeds of the plant and the number of viable pods, results in grams (g). Number of branches (NB) was counted of the number of branches larger than ten centimeters. Mass of seeds per plant (MSPP) was the pods harvested on each plant were threshed, subsequently it was determined the humidity degree and the mass of seeds was adjusted to 13% moisture. Seed length (SL) was measured by the length of all plant seeds, character determined by using a digital caliper. Seed width (SW) was measured by the width of all plant seeds, character determined through a digital caliper. Seed flattening (SF) was determined by the methodology proposed by Silva (2005), wherein seeds are classified as flattened, semi-flattened and filled. Seed Brightness (BRI) was conducted by visual grading through the methodology of Silva (2005), wherein seeds are classified as opaque, intermediate and bright. Presence of halo (HAL) was visual assessed determining the presence or absence of halo in the seed. Color of the halo (CRH) was visual assessment determining the color of the halo in relation to the seed.

Statistical analysis

The data obtained were submitted to descriptive analysis. Thereafter, the distributions of phenotypic frequencies of the traits were performed. Analysis of genetic dissimilarity between genotypes by standardized Euclidean distance was subsequently carried out, applying the UPGMA (unweighted pair grouping method with arithmetic mean). The relative importance of the characters was obtained by the methodology proposed by Singh (1981). The Tocher optimization method was held based on the matrix of the standardized Euclidean distance, and then the principal component analysis was held. All analyses were performed using the Genes software (Cruz, 2013).

Conclusions

The distributions of phenotypic frequencies indicate genetic variability among the 149 genotypes of common black bean. The Tocher optimization method presents the formation of eight groups of genotypes. The dendrogram formed by the standardized Euclidean distance was efficient in the stratification of accessions for their genetic distance. The relative contribution by Singh method shows that days to flowering and seed brightness are the characters that best discriminate genotypes. The multivariate techniques of Tocher optimization and standardized Euclidean distance show similar responses, proving to be viable tools for the choice of parents in a genetic breeding program.

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