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Genetic parameters and combined selection for seed coat color and macrominerals in Mesoamerican common bean lines

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ABSTRACT. The development of common bean cultivars with biofortified grains for macrominerals and a seed coat color pattern that satisfies consumer preference is a new demand of breeding programs. However, combined selection for seed coat color and macromineral concentrations in common beans is unprecedented. The objectives of this work were to obtain estimates of genetic parameters for seed coat color and macromineral concentrations in a recombinant inbred line population of Mesoamerican common beans and to select superior common bean lines for a biofortification program. A biparental cross BRS Esteio × SCS 205 Riqueza resulted in 100 Mesoamerican common bean lines that were evaluated in the $F_{5:6}$ and $F_{5:7}$ generations. Seed coat color was evaluated in a colorimeter by values L* (white to black), a* (green to red), and b* (blue to yellow). Macromineral concentrations (potassium, calcium, and magnesium) were quantified by nitric-perchloric acid digestion. A significant treatment effect for all traits evaluated at the $F_{5:6}$ and $F_{5:7}$ generations was observed. Seed coat color (L*, a*, and b* values) presented high heritability estimates (h^2: 95.99 to 99.72%) and qualitative inheritance. Heritability estimates from intermediate to high magnitude (h^2: 34.08 to 99.50%) were obtained for the concentrations of potassium, calcium, and magnesium. Transgressive segregation and quantitative inheritance for the macromineral concentrations were observed in the $F_{5:7}$ generation. The four superior common bean lines selected by the $\overline{Z}$ index were L 83-17, L 69-17, BRS MG Pioneiro, and L 24-17. The lines L 83-17 and L 69-17 of black beans have darker grains (lower L*, a*, and b* values) and high concentrations of potassium, calcium, and magnesium. The lines BRS MG Pioneiro and L 24-17 of carioca beans present lighter grains (L* ≥ 55, a* ≤ 7 and b* ≤ 16), and high mineral concentration. These lines will be selected for a common bean biofortification program.

Key words: *Phaseolus vulgaris;* Heritability; Inheritance pattern; Selection index

INTRODUCTION

Beans (*Phaseolus vulgaris*) are an extremely healthy food, consumed by people from diverse social classes, in many countries. That is due to its high nutritional value and the health benefits associated with regular consumption of beans (Messina, 2014; Suárez-Martínez et al., 2016; Chávez-Mendoza and Sánchez, 2017). For this reason, beans are a promising crop for genetic biofortification programs.

Bean biofortification has focused on the increase of iron and zinc concentrations, aiming to prevent the deficiency of these minerals that constitute a public health problem (Blair, 2013; Petry et al., 2015). Biofortification for macrominerals is recent in common beans. Breeding programs developed Mesoamerican common bean lines with high concentrations of potassium (Maziero et al., 2016), calcium (Casañas et al., 2013; Ribeiro et al., 2014), and magnesium (Casañas et al., 2013). Potassium, calcium, and magnesium are the largest cations found in beans (Suárez-Martínez et al., 2016). These macrominerals are responsible for major functions in the human organism. Potassium helps lower blood pressure and reduces diabetes risk (Ekmekcioglu et al., 2016). Calcium is essential for the formation and maintenance of bones, and skeletal integrity (Wimalawansa et al., 2018). Magnesium helps prevent diabetes, osteoporosis, and cardiovascular diseases (Alawi et al., 2018).

The development of common bean cultivars with biofortified grains for macrominerals meets a world-wide demand for foods with high nutritional value. Thus, the study of the genetic parameters of macrominerals is of the utmost importance. The heritability of the potassium concentration in Mesoamerican common beans ranged from low to intermediate magnitude (h^2: 28.80 to 59.10%) at the $F_{6:8}$ generation (Maziero et al., 2016), and the calcium concentration had high heritability (h^2: 64.78 to 66.85%) at the F_7 generation (Ribeiro et al., 2014). Heritability estimates for magnesium concentration in a recombinant inbred line population of common bean were not found in published reports. A study of genetic parameters for the concentrations of potassium, calcium, and magnesium in a single recombinant inbred line population of Mesoamerican common bean is also unprecedented.

The development and launch of a new common bean cultivar biofortified for macrominerals can only be successful if the seed coat color is accepted by consumers. Common bean seed coat color can be evaluated qualitatively by visual observation, or quantitatively by the use of a colorimeter. The colorimeter enables one to differentiate the seed coat color using three different parameters: L* characterizes the luminosity (dark to light); a* evaluates the variation of the green to red shades; and b* measures the variation of the blue to yellow shades. These parameters have shown to be effective for the selection of common bean lines with seed coat color pattern that satisfy consumer preference of the black and carioca beans (Ribeiro et al., 2003, 2008; Possobom et al., 2015; Arns et al., 2018). However, the combined selection for seed coat color quantitatively evaluated by L*, a*, and b* values and macromineral concentrations (potassium, calcium, and magnesium) in Mesoamerican common bean is still unprecedented. This can enable the development of common bean cultivars biofortified for macrominerals and with a seed coat color pattern that is accepted by consumers; this is a new demand for breeding programs.

Thus, the objectives of this work were to obtain estimates of genetic parameters for seed coat color and macromineral concentrations in a recombinant inbred lines population of Mesoamerican common bean, and to select superior common bean lines for a biofortification program.

MATERIAL AND METHODS

Obtaining of the recombinant inbred lines

A population of 100 recombinant inbred lines was obtained from crosses between Mesoamerican common bean cultivars BRS Esteio and SCS 205 Riqueza. BRS Esteio has black grains, and was originally denominated as line CNFP 10104 by the Brazilian Agricultural Research Corporation (EMBRAPA). SCS 205 Riqueza has carioca grains (beige seed coat with brown streaks), and was originally denominated as line CHC 01-175 by the Agricultural Research and Rural Extension Corporation (EPAGRI). BRS Esteio and SCS 205 Riqueza have different concentrations of potassium and calcium in the grains (Ribeiro et al., 2013a), and represent the most consumed common bean types in Brazil, therefore they were chosen as parents.

The emasculation of floral bud method with the interlacing of the stigmas of both parents was used to obtain the F_1 seeds, in 2012 (Possobom et al., 2015). The recombinants obtained were advanced up to the F_5 generation using the Single-Seed Descent method. The seeds obtained from each plant of the F_5 generation were individually harvested for the evaluation of the common bean lines in field experiments.

Evaluation of the $F_{5:6}$ and $F_{5:7}$ generations and determination of the seed coat color and macromineral concentrations

Two experiments were carried out in an area of the Bean Breeding Program of the Federal University of Santa Maria, Santa Maria, Rio Grande do Sul, Brazil (latitude 29°42’S, longitude 53°43’W, and altitude 95 m). The first experiment was sown on March 09, 2017 (rainy season) and the second experiment was sown on October 28, 2017 (dry season). Rainy and dry seasons characterize the traditional growing seasons for common bean in the South of Brazil.

The region has a humid subtropical climate, with hot summers and without a clearly defined dry season. The soil is classified as a typical alitic Argisol, Hapludalf, and presented the following chemical composition: 18 g.kg^{-1} organic matter; 73.90% of base saturation; 6.10 of pH (H_20); 9.90 cmolc.dm^{-3} of effective cation exchange capacity; 80.00 mg.dm^{-3} potassium; 12.70 mg.dm^{-3} phosphorus; 6.70 cmolc.dm^{-3} calcium; 2,026.50 mg.dm^{-3} iron; 1.20 mg.dm^{-3} copper; and 0.50 mg.dm^{-3} zinc. The soil was conventionally prepared and the amount of fertilizers was calculated based on the interpretation soil chemical analysis.

The first experiment was performed in an augmented block design, with two replications. Treatments consisted of 103 common bean genotypes, with 100 $F_{5:6}$ lines, two parent cultivars (BRS Esteio and SCS 205 Riqueza), and one control cultivar (Pérola). The second experiment was carried out in a 11 x 11 simple lattice design and was composed of 121 common bean genotypes, with 100 $F_{5:7}$ lines, two parent cultivars, and 19 control cultivars. The experimental plot consisted of one 1 m row, spaced 0.50 m apart in the two experiments. The sowing density used was 15 seeds per linear meter.

Similar management practices were used on both experiments. Seeds were treated at sowing with Maxim ML fungicide (Fludioxonil and Metalaxyl-M) and Cruiser insecticide (Thiamethoxam), both at a dose of 200 mL/100 kg of seeds. During the development of the crop, Engeo™ Pleno insecticide was applied (Thiamethoxam and Lambda-cyhalothrin) at a dose of 125 mL/ha whenever the infestation of *Diabrotica speciosa* caused a reduction of approximately 5% of the leaf area. Weeds were mechanically removed every 10 days, in order to avoid competition with the crop.

Plants were harvested when pods lost their pigmentation and began to dry up and grains presented the typical color of the genotype (R9 stage). Processing was performed manually, and the beans were dried on an oven (Odontobras 1.5; Odontobras, São Paulo, Brazil) at 40°C, until 13% of moisture. The seed coat color was evaluated with a portable colorimeter (CR 410, Konica Minolta, Osaka, Japan). Reflectance was measured using the CIE L* a* b* color scale in grain samples placed on a petri dish of 6.0 cm diameter and 1.5 cm height. The L* parameter defined the luminosity of the sample and ranges from light to dark (0 to 100); the chromaticity a* value evaluated the variation of the green to red shades (-60 to 60); and the chromaticity b* value measured the variation of the blue to yellow shades (-60 to 60). The readings were taken in triplicate for each experimental plot.

Grain samples of 10 g were randomly collected from each experimental plot for the macromineral analysis. The grains were ground in an analytical micro-mill (Q298A21, Quimis, São Paulo, Brazil) to obtain particles smaller than 1 mm in diameter, not sieved. Aliquots of 0.5 g of the flour obtained were used for the nitric-perchloric acid digestion (HNO_3 + $HClO_4$, in a 3:1 ratio by volume), as described by Miyazawa et al. (2009). The potassium concentration was obtained by a flame photometer (B262, Micronal, São Paulo, Brazil), with a wavelength of 660.0 nm. The calcium and magnesium concentrations were determined by an atomic absorption spectrometer (AAS, Perkin Elmer AAnalyst 200, Waltham, United States of America), with a wavelength of 422.7 nm and 285.2 nm, respectively.

Estimates of genetic parameters and selection index

Data obtained were subjected to analysis of variance, according to the augmented block ($F_{5:6}$ generation) and simple lattice ($F_{5:7}$ generation) designs. The efficiency of the simple lattice design, compared to a randomized block design was determined as described by Maziero et al. (2016).

The estimates of genetic parameters were obtained for each trait evaluated at the $F_{5:6}$ and $F_{5:7}$ generations, namely: phenotypic variance, environmental variance, genetic variance, heritability, coefficient of experimental variation (CEV), coefficient of genetic variation (CGV), relationship between the CGV and the CEV (CGV/CEV ratio), and selective accuracy. The heritability was obtained based on the variance components, using the means of treatments, by the formula:

$$h^2 = \frac{\sigma^2 G}{\sigma^2 F} \quad \text{(Eq. 1)}$$

were: σ^2G is the genetic variance, and σ^2F is the phenotypic variance. The heritability determined for the $F_{5:6}$ and $F_{5:7}$ generations corresponds to narrow-sense heritability, since homozygosis is high in these generations. Thus, the genetic variance corresponds to the additive variance.

Data normality was verified by the Shapiro-Wilk test ($P < 0.05$). Frequency distribution graphs for each trait evaluated were obtained at the $F_{5:6}$ and $F_{5:7}$ generations. In each graph, the number of classes was established by the expression $\sqrt{n}$, where n is the number of observations.

The selection index ($\overline{Z}$ index) was estimated as described by Mendes et al. (2009). The mean of the $\overline{Z}$ index obtained for the traits evaluated at the $F_{5:7}$ generation was added to a constant equal to three to avoid negative values. The contribution of each standardized trait in the value of the $\overline{Z}$ index was presented in charts for the three superior common bean lines of black and carioca beans. The charts were made in Microsoft Office Excel spreadsheets, and the statistical analyses were carried out using Genes software (Cruz, 2016).

RESULTS AND DISCUSSION

Genetic variability and genetic parameters for seed coat color in Mesoamerican common beans

A significant treatment effect for the seed coat color (L*, a*, and b* values) was observed at the $F_{5:6}$ and $F_{5:7}$ generations (Table 1). Therefore, there is genetic variability among the common bean lines evaluated for the seed coat color, which varied from dark to light (L* value), and presented variations in the green-red (a* value) and blue-yellow (b* value) shades (Figure 1A, 1B, 1C, 1D, 1E, and 1F). Recombinant inbred lines with great variation for the L*, a*, and b* values were previously obtained from controlled crosses between the common bean parents that had differences in the seed coat color (Ribeiro et al., 2009; Erfatpour et al., 2018). Thus, the use of colorimeter to determine the seed coat color allows the differentiation of common bean lines within and between different commercial groups obtained by the breeding programs.

The values of coefficient of experimental variation ≤20.70% and selective accuracy ≥0.980 (Table 1) showed that the L*, a*, and b* values were evaluated with high experimental precision. Arns et al. (2018) also observed high experimental precision when determining L*, a*, and b* values in carioca common bean lines. The lower contribution of the environmental in the expression of the seed coat color in common bean can explain the results obtained. As a consequence, the selection of common bean lines based on lightness grains, when based on L* value, can be performed in a single experiment with an accuracy of 99% (Cargnelutti et al., 2006).

Table 1. Analysis of variance and genetic parameters estimates for seed coat color (L*, a*, and b* values) and the concentrations of potassium, calcium, and magnesium obtained in the Mesoamerican common bean lines at the $F_{5:6}$ and $F_{5:7}$ generations.

Sources of variance	**DF**[1]	**Mean square** L*	a*	b*	K	Ca	Mg
					... g kg^{-1} of dry matter ...		
	$F_{5:6}$ generation						
Block	1	115.90	3.57	45.84	223.63	0.18	0.00
Treatment (adjusted)	102	227.37*	5.17*	65.99*	3.77*	0.06*	0.10*
Residue	2	1.12	0.05	0.89	0.03	0.00	0.00
General mean		34.27	2.95	7.29	13.58	1.30	2.71
Common mean – control		46.47	2.75	11.19	13.15	1.23	2.70
Non-common mean – lines		33.53	2.97	7.05	13.60	1.30	2.71
Phenotypic variance		207.959	5.281	64.429	6.647	0.061	0.104
Environmental variance		1.118	0.054	0.893	0.033	0.001	0.005
Genotypic variance		206.842	5.227	63.536	6.611	0.061	0.098
Heritability (%)		99.46	98.98	98.61	99.50	98.94	94.84
CEV (%)[2]		3.15	7.83	13.40	1.34	1.95	2.70
CGV (%)[3]		42.89	77.08	113.03	18.90	18.90	11.56
CGV/CEV ratio[4]		13.60	9.84	8.44	14.12	9.68	4.29
Selective acuracy		0.998	0.995	0.993	0.996	0.994	0.974
	$F_{5:7}$ generation						
Replication	1	2.79	2.92	3.57	1.14	1.35	0.08
Block/replication (ajusted)	20	2.44	0.43	0.39	1.48	7.67	2.40
Treatment (ajusted)	120	517.61*	9.48*	128.62*	1.11*	0.11*	0.01*
Residue	100	1.77	0.38	0.35	0.65	0.07	0.00
Lattice efficiency (%)		102.20	100.42	100.29	113.81	100.45	91.70
General mean		35.33	2.98	7.83	11.07	2.02	1.35
Control mean		40.69	3.20	9.31	11.25	1.92	1.34
Lines mean		34.21	2.93	7.52	11.04	2.04	1.35
Phenotypic variance		258.805	4.738	64.310	0.557	0.055	0.004
Environmental variance		0.887	0.189	0.176	0.325	0.033	0.003
Genotypic variance		257.918	4.549	64.134	0.232	0.022	0.001
Heritability (%)		99.66	95.99	99.72	41.73	40.03	34.08
CEV (%)[2]		3.77	20.70	7.58	7.28	12.79	5.11
CGV (%)[3]		45.45	71.64	102.24	4.35	7.39	2.60
CGV/CEV ratio[4]		12.06	3.46	13.48	0.60	0.58	0.51
Selective acuracy		0.998	0.980	0.999	0.646	0.633	0.584

[1]Degree freedon. [2]Coefficient of environmental variation. [3]Coefficient of genetic variation. [4]Coefficient of genetic variation and coefficient of environmental variation ratio. *Significant by F test at 0.05 probability.

High heritability estimates were obtained for the L*, a*, and b* values (h^2: 95.99 to 99.72%) at the $F_{5:6}$ and $F_{5:7}$ generations. Ribeiro et al. (2009) also observed that the L* value showed high heritability (h^2: 91.13%) in common bean lines at the $F_{6:7}$ generation obtained from 11 different hybrid combinations. Nevertheless, for common bean no previous studies of heritability estimates for the a* and b* values in generation of high homozygosis were found in the literature. In segregating generation, Possobom et al. (2015) verified high heritability for the L*, a*, and b* values in recombinants of Mesoamerican and Andean common bean. High heritability estimates are associated with the predominance of genetic variance. In addition, most of the genetic variance is attributed to additive variance in generations with high homozygosis in the autogamous plants, such as common bean (Ramalho et al., 2012). The additive variance can be fixed, thus the selection of common bean lines with superior L*, a*, and b* values at the $F_{5:7}$ generation obtained from the crosses between BRS Esteio and SCS 205 Riqueza is expected to be easier.

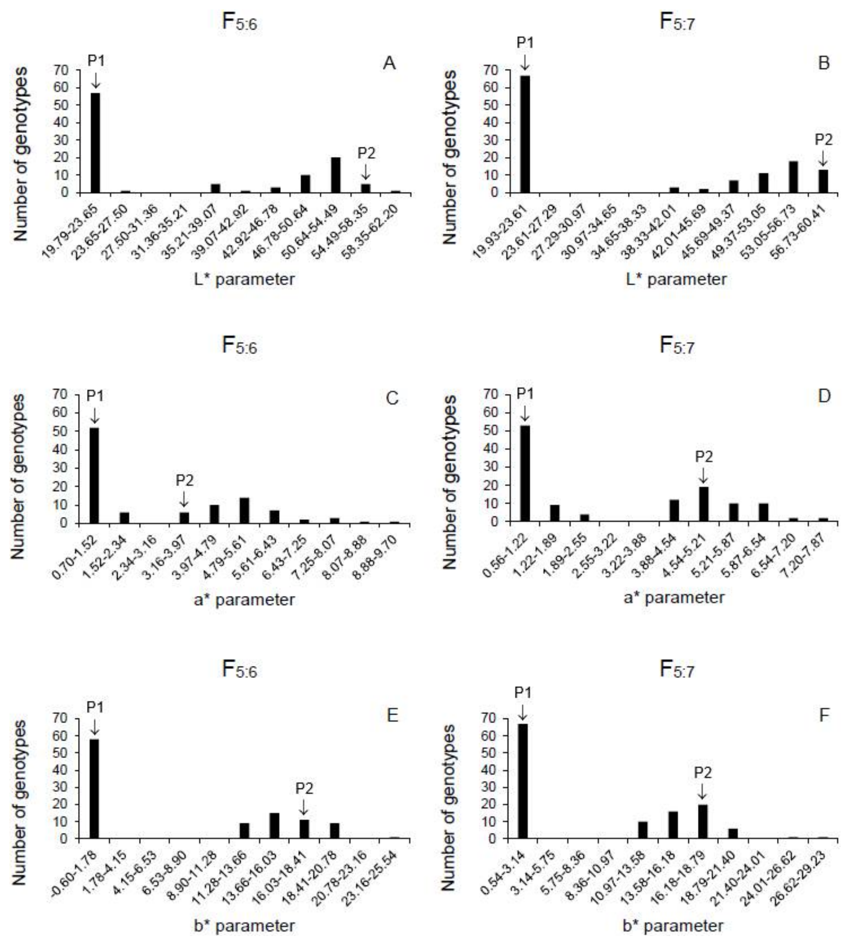

Figure 1. Frequency distribution for the seed coat color (L*, a*, and b* values) in Mesoamerican common bean lines at the $F_{5:6}$ and $F_{5:7}$ generations. P1 and P2 are parent cultivars: P1 (BRS Esteio) and P2 (SCS 205 Riqueza).

High coefficients of genetic variation for the L*, a*, and b* values (42.89 to 113.03%) were obtained, indicating little interference from the environment on these traits. Ribeiro et al. (2008) also found high coefficient of genetic variation for the L* value determined in experiments that assessed Mesoamerican common bean lines. Furthermore, the CGV/CEV ratio, that represents the relationship between the coefficient of genetic variation and the coefficient of experimental variation, was high (≥3.46) for the L*, a*, and b* values, in the present study. According to Jost et al. (2013), when the CGV/CEV ratio values are close or superior to the unit, there are greater possibilities of gain with the selection. High coefficient of genetic variation and CGV/CEV ratio values observed in the present study indicate facilities for the selection of genetically superior common bean lines based on the L*, a*, and b* values.

The L*, a*, and b* values presented discontinuous variation at the $F_{5:6}$ and $F_{5:7}$ generations (Figure 1A, 1B, 1C, 1D, 1E, and 1F), indicating that few genes act in the manifestation of the seed coat color in common bean. Similarly, Possobom et al. (2015) observed the formations of few phenotypic classes for the L*, a*, and b* values determined in recombinants of Mesoamerican and Andean common bean at the F_2 generation, which characterized a qualitative inheritance. The genetic control of the darkening of carioca beans was described as governed by two genes with recessive epistatic interaction (Silva et al., 2014; Erfatpour et al., 2018). In the present study, the results of frequency distribution obtained at the $F_{5:6}$ and $F_{5:7}$ generations allow to infer that L*, a*, and b* values present qualitative inheritance, which facilitates the selection of common bean lines with seed coat color that have acceptance of the consumer.

Genetic variability and genetic parameters for the macromineral concentrations in Mesoamerican common bean

A significant treatment effect for the concentrations of potassium, calcium, and magnesium was verified in the analysis of variance at the $F_{5:6}$ and $F_{5:7}$ generations (Table 1), indicating that there is genetic variability for the macromineral concentrations among Mesoamerican common bean lines. Previous studies showed that was possible to develop common bean lines with high concentrations of potassium (Maziero et al., 2016), calcium (Casañas et al., 2013; Ribeiro et al., 2014), and magnesium (Casañas et al., 2013) from controlled crosses between parents with contrasting macromineral concentrations. These results evidence that it is possible to launch common bean cultivars with biofortified grains for macrominerals, meeting a world-wide demand for foods with high nutritional value.

The coefficient of experimental variation ranged from 1.34 to 12.79%, which are lower values then the ones previously described by Maziero et al. (2016) and Ribeiro et al. (2014) for the potassium and calcium concentrations assessed in recombinant inbred line populations of Mesoamerican common bean. Similarly, estimates of coefficient of experimental variation ≤12.00% were obtained for concentrations of potassium, calcium, and magnesium evaluated in a collection of 277 common bean genotypes from Mesoamerican gene pool by McClean et al (2017). This allows us to infer that the concentrations of potassium, calcium, and magnesium were evaluated with high experimental precision in our study, when considering the coefficient of experimental variation. Also, the selective accuracy revealed very high experimental precision ($SA \geq 0.90$) in determining the macrominerals at the $F_{5:6}$ generation and intermediate ($0.50 \leq SA < 0.65$) at the $F_{5:7}$ generation, considering the classes established by Resende and Duarte (2007). High coefficient of experimental variation and lower selective accuracy values were observed at the $F_{5:7}$ generation, resulting in a lower experimental precision in the evaluation of the macrominerals. This can be an indicative of higher contribution of the environmental on the expression of the concentrations of potassium, calcium, and magnesium at the $F_{5:7}$ generation.

The concentrations of potassium, calcium, and magnesium presented high heritability ($h^2 \geq 94.84\%$) at the $F_{5:6}$ generation, and intermediate heritability ($34.08 \leq h^2 < 41.73\%$) at the $F_{5:7}$ generation in Mesoamerican common bean lines. Maziero et al. (2016) observed that the heritability of the potassium concentration ranged from low to intermediate magnitude in the four recombinant inbred line populations of Mesoamerican common bean at the $F_{6:8}$ generation. Ribeiro et al. (2014) verified a high heritability estimative for calcium concentration in Mesoamerican common bean lines at the F_7 generation, regardless of the method used to advance of the segregating generations. High broad-sense heritability was found for the concentrations of potassium, calcium, and magnesium evaluated in a collection of 277 common bean genotypes that includes old landraces, cultivars and advanced generations breeding lines from several breeding programs in the United States of America (McClean et al., 2017). Thus, the heritability of the macromineral concentrations in Mesoamerican common bean is not a fixed trait, it can vary with the generation and the recombinant inbred line population assessed, and is dependent of the genetic diversity of the parents used in the crossings. In the present study, a higher contribution of the environmental variance in the expression of the phenotypic variance was observed at the $F_{5:7}$ generation, which resulted in lower heritability value.

Relatively high values (2.60 to 18.90%) for the coefficient of genetic variation were obtained for concentrations of potassium, calcium, and magnesium from both generations assessed, which indicates low uncontrollable influence in the expression of these traits. However, CGV/CEV ratio values were higher than the unit only at the $F_{5:6}$ generation, which suggests a more favorable scenario for the selection of common bean lines biofortified in this generation. Conversely, CGV/CEV ratio values lower than the unit are not indicative of an unsuccessful selection. Maziero et al. (2016) verified relatively low CGV/CEV ratio values (0.30 to 0.60) for the potassium concentration, but common bean lines biofortified were obtained in the four recombinant inbred line populations.

The concentrations of potassium, calcium, and magnesium did not present a normal distribution at the $F_{5:6}$ generation when evaluated by the Shapiro-Wilk test (Figure 2A, 2C, and 2E). However, these macrominerals presented a continuous distribution at the $F_{5:7}$ generation, nearly normal distribution, indicating quantitative inheritance (Figure 2B, 2D, and 2F). McClean et al. (2017) observed that potassium, calcium, and magnesium were normally distributed when they evaluated 277 common bean genotypes of the most important market classes cultivated in the United States. In F_7 lines obtained from crosses between Mesoamerican common bean parents, a continuous distribution for calcium concentration in the grains was also verified (Ribeiro et al., 2014). Similarly, a continuous distribution was observed for the calcium and magnesium concentrations in a recombinant inbred line population obtained from the cross between Mesoamerican and Andean common bean cultivars (Casañas et al., 2013). Katuuramu et al. (2018) determined the potassium and calcium concentrations in cooked grains of 206 genotypes of Andean common bean and verified a normal distribution for these macrominerals. These results indicate that the selection for the concentrations of potassium, calcium, and magnesium can be more difficult due to the effect of individual locus influencing these traits, which are difficult to isolate and characterize. However, the assisted selection with molecular markers can be a viable alternative to the biofortification programs in common beans. Recent studies revealed the discovery of two quantitative trait loci (QTL) associated with the potassium concentration in a recombinant inbred line of common bean ($F_{9:11}$ generation), one QTL was found for calcium, and three QTL´s were found for magnesium (Blair et al., 2016).

In the $F_{5:7}$ generation were obtained Mesoamerican common bean lines with macromineral concentrations ranging from 9.24 to 12.81 g/kg of dry matter (DM) for potassium (Figure 2B); from 1.48 to 2.62 g/kg DM for calcium (Figure 2D); and from 1.21 to 1.57 g/kg DM for magnesium (Figure 2F). Superior values for the potassium (Maziero et al., 2016) and magnesium (Casañas et al., 2013) concentrations, and similar value for the calcium concentration (Ribeiro et al., 2014) were observed in recombinant inbred lines of common bean. In the present study, were obtained Mesoamerican common bean lines of black and carioca grains with superior and inferior concentrations of potassium, calcium, and magnesium to the values obtained by the parents, characterizing a transgressive segregation. Transgressive segregation for the concentrations of potassium, calcium, and magnesium in common bean lines has been previously described by Maziero et al. (2016), Ribeiro et al. (2014), and Casañas et al. (2013), respectively. Transgressive segregation could be mainly explained by complementary gene action or epistasis, according by Casañas et al. (2013), and recombinant events would have generated new genotypes, resulting in phenotypes different to the parents.

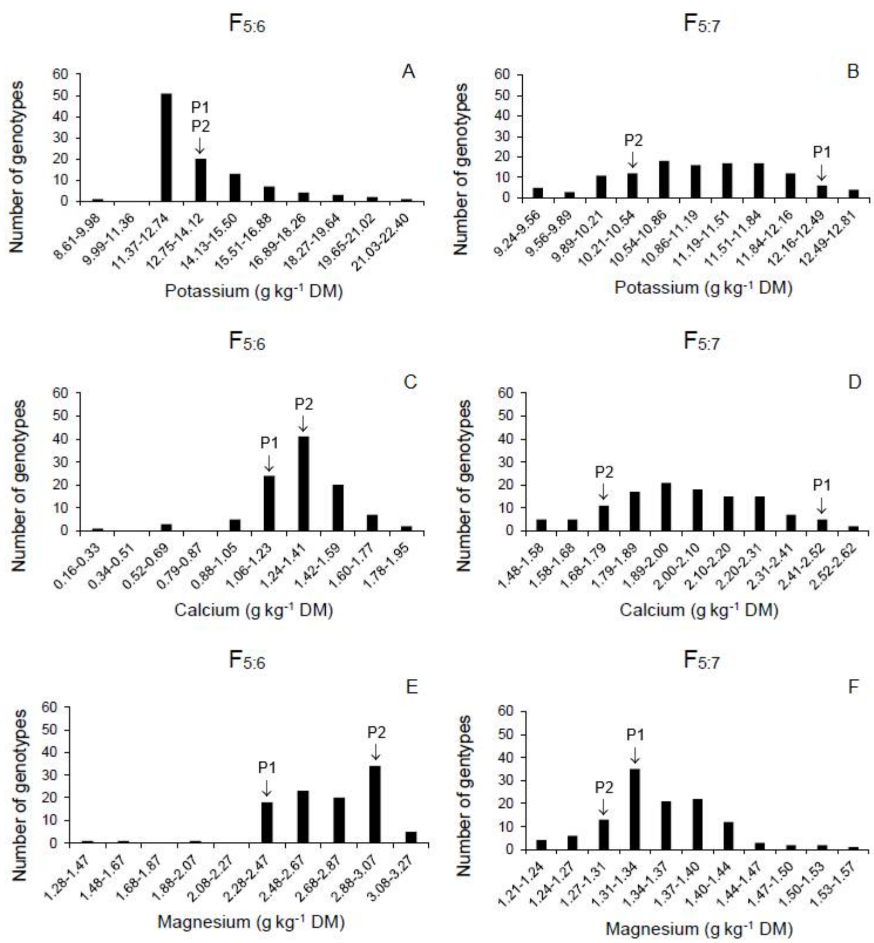

Figure 2. Frequency distribution for the concentrations of potassium, calcium, and magnesium obtained in the Mesoamerican common bean lines at the $F_{5:6}$ and $F_{5:7}$ generations. P1 and P2 are parent cultivars: P1 (BRS Esteio) and P2 (SCS 205 Riqueza).

When transgressive segregation is observed in a crossing, it is possible to select recombinants with high macromineral concentrations. In this study, 12 Mesoamerican common bean lines presented high potassium concentration, i.e., ≥ 12 g/kg DM (Figure 2B), as previously established by Steckling et al. (2017). All common bean lines obtained have a high calcium concentration (≥1.48 g/kg DM, Figure 2D), considering the classes established for common bean by Ribeiro et al. (2013b). No previous studies were found in the literature determining which characterize high and low values for magnesium concentration in common bean. However, considering the previously described values for magnesium in common bean lines (Silva et al., 2012; Casañas et al., 2013), it is possible to say that the magnesium concentration was low for most of the $F_{5:7}$ lines evaluated in the present study.

Combined selection for seed coat color and macrominerals in Mesoamerican common bean

The use of the $\overline{Z}$ selection index made possible the graphical display and the selection of superior Mesoamerican common bean lines in regarding the seed coat color (L*, a* and, b* values) and macromineral concentrations (potassium, calcium, and magnesium). For black beans, the three superior lines selected were L 83-17, L 69-17, and L 16-17 (Figure 3A, 3B, and 3C). These lines showed the lowest means of the $\overline{Z}$ index for the L*, a*, and b* values. Common bean lines of black grains must have a L* value ≤ 22, because this indicates the absence of purple grains (Ribeiro et al., 2003), and are associated with fresh and fast cooking grains by consumers. In addition, a* and b* values must be close to zero, indicating the absence of a secondary color (Possobom et al., 2015). The lines L 83-17, L 69-17, and L 16-17 have L*, a*, and b* values that meet the standard for seed coat color required by breeding programs for black beans. Furthermore, these three lines showed the highest $\overline{Z}$ index for the potassium and calcium concentrations. The lines L 83-17 and L 69-17 of black beans had darker grains (lower L*, a*, and b* values) and presented biofortified grains for potassium, calcium, and magnesium, thus having a higher nutritional value. Conversely, McClean et al. (2017) did not find high concentrations of potassium, calcium, and magnesium in black bean genotypes when they evaluated a collection of 277 Mesoamerican genotypes of different market classes. Black bean genotypes from a Mesoamerican gene pool have been associated with high concentrations of sulfur, manganese, and zinc (Hacisalihoglu and Settles, 2013).

For carioca beans, the breeding programs have been prioritizing the selection of lighter grains, i.e., L* value ≥55 (Ribeiro et al., 2008), because consumers associate the lighter color with fresh and fast cooking grains. Additionally, carioca beans must have an a* value ≤7 (slightly red shade), and a b* value ≤16 (slightly yellow shade), because these combination of colors results in lighter grains and of slow darkening (Arns et al., 2018). The three superior lines of carioca beans selected by the $\overline{Z}$ index were BRS MG Pioneiro, L 61-17, and L 24-17 (Figure 3D, 3E, and 3F). BRS MG Pioneiro had the highest means of the $\overline{Z}$ index for the L* value, concentrations of potassium, calcium, and magnesium, and the lowest $\overline{Z}$ index for the a* and b* values. BRS MG Pioneiro meets the demand of the consumers for lighter grains and of low darkening for carioca beans and had biofortified grains for macrominerals. L 61-17 attends the standards required for seed coat color for carioca beans and showed biofortified grains for potassium and calcium. L 24-17 had a $\overline{Z}$ index close to the mean for L* value, i.e., the grains were slightly darker than those of the lines BRS MG Pioneiro and L 61-17, but still with grains standard that meet the demand of the consumers. L 24-17 also had higher $\overline{Z}$ index for the concentrations of potassium, calcium, and magnesium. Similarly, Morais et al. (2016) selected common bean landraces and cultivars with high mineral concentrations using the $\overline{Z}$ index.

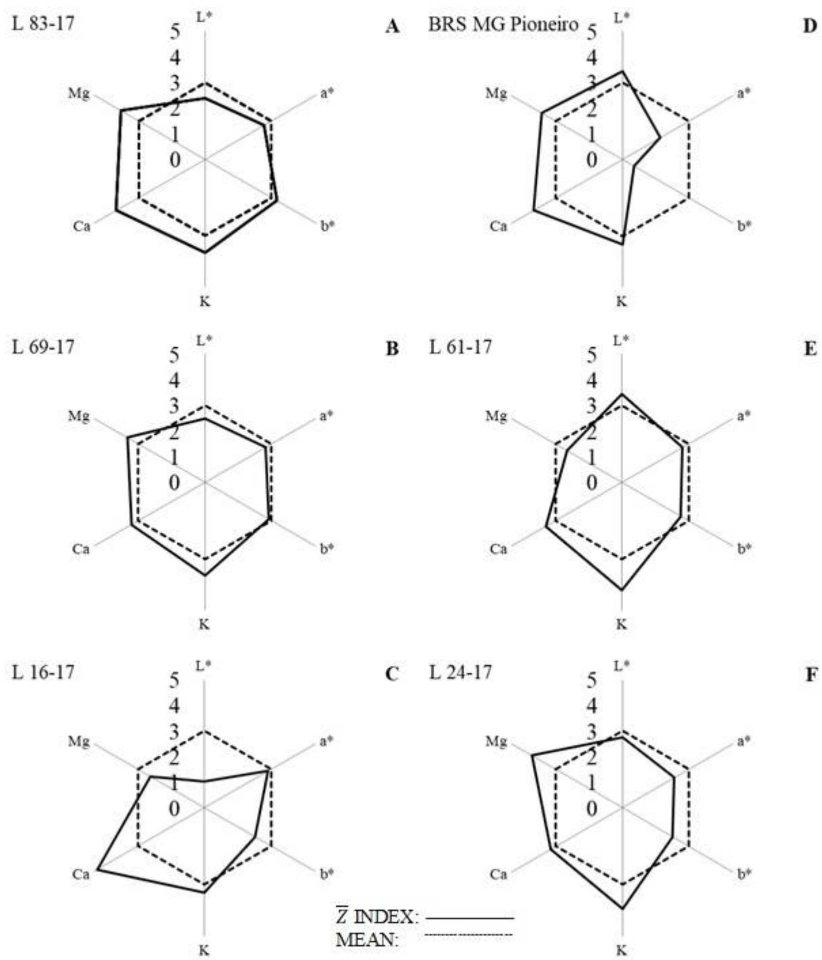

Figure 3. Representation of the $\overline{Z}$ index for luminosity (L*), chromaticity a* (a*), chromaticity b* (b*), concentrations of potassium (K, $g.kg^{-1}$ of dry matter - DM), calcium (Ca, $g.kg^{-1}$ DM), and magnesium (Mg, $g.kg^{-1}$ DM) obtained from three superior common bean lines of black beans (A, B, and C) and carioca beans (D, E, and F).

The Mesoamerican common bean lines of black beans L 83-17 and L 69-17 and of carioca beans BRS MG Pioneiro and L 24-17 presented seed coat color that satisfy consumer preference, and had biofortified grains for potassium, calcium, and magnesium. Diets rich in potassium can lower blood pressure, especially in hypertensive individuals, and reduce diabetes risk (Ekmekcioglu et al., 2016) and metabolic syndrome (Lee et al., 2013). Calcium is necessary to the formation and maintenance of bones and skeletal integrity, thus it is essential to prevent osteoporosis (Wimalawansa et al., 2018). Magnesium plays an important function in preventing diabetes, osteoporosis, migraines, and cardiovascular diseases (Alawi et al., 2018). The lines L 83-17, L 69-17, BRS MG Pioneiro, and L 24-17 if consumed regularly as part of a balanced diet can represent health benefits due the highest concentrations of potassium, calcium, and magnesium. In addition, these lines had seed coat color that satisfy consumer preference. The lines L 83-17, L 69-17, BRS MG Pioneiro, and L 24-17 will be selected for the common bean biofortification program.

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CONFLICTS OF INTEREST

The authors declare no conflict of interest.

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