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Phenotypic diversity of nutrients and anti-nutrients in bean grains grown in different locations

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ABSTRACT

The growing environments can influence grain yield and could also to influence the nutritional value of common bean grains. The objective of this study was to determine the levels of minerals, phytic acid and total protein in common bean, and to the relationship of these with grain yield, under the influence of different growing environments. The experiment was conducted with 26 genotypes in three Santa Catarina places of cropping (Anchieta/SC, Joaçaba/SC and Lages/SC, Brazil). Were determined the levels of phosphorus, potassium, iron, zinc, protein and phytic acid into grains. The levels of phytate, iron and potassium, considering each genotype were influenced by the environment of cropping while for zinc, phosphorus and crude protein there was no interaction of genotypes by environments effects. In addition, there was variability for all nutrients and phytic acid in at least one cropping place. Genotypes with high grain yield had lower concentrations of nutrients, particularly phosphorus and crude protein, although this relationship is dependent of the cropping place. Concomitant choice for two minerals was possible; however, there was a positive correlation between mineral and phytate which was not favorable because the nutrients complexes with phytic acid, characterizing phytic acid as an important grains anti-nutrient.

Key words: phytate; minerals; *Phaseolus vulgaris* L.; crude protein

Diversidade fenotípica de nutrientes e anti-nutriente em grãos de feijão cultivados em diferentes locais

RESUMO

O ambiente de cultivo influencia no rendimento de grãos e pode também influenciar o valor nutricional de grãos de feijão. O objetivo deste trabalho foi determinar os teores de minerais, proteína total e ácido fítico em grãos de feijão e, a relação destes com o rendimento de grãos, sob a influência de diferentes ambientes de cultivo. O experimento foi conduzido com 26 genótipos de feijão em três locais de cultivo (Anchieta/SC, Joaçaba/SC e Lages/SC) na safra 2008/2009, onde se determinaram os teores de fósforo, potássio, ferro, zinco, proteína bruta e ácido fítico nos grãos e o rendimento de grãos. Os genótipos, quanto aos teores de ácido fítico, ferro e potássio, foram influenciados pelo local de cultivo enquanto, para zinco, fósforo e proteína bruta não houve interação dos genótipos com os ambientes. Em adição, observou-se variabilidade genotípica para todos os nutrientes e o ácido fítico em pelo menos um local de cultivo. Os genótipos com elevado rendimento de grãos apresentaram menores concentrações de nutrientes nos grãos, principalmente fósforo, assim como de proteína bruta, porém essa relação é dependente do local de cultivo. A escolha concomitante para dois minerais foi possível, entretanto, também houve correlação positiva entre os minerais e ácido fítico o que não foi favorável, pois os nutrientes estão complexados com ácido fítico, caracterizando-o como um anti-nutriente importante nos grãos.

Palavras-chave: fitato; minerais; *Phaseolus vulgaris* L.; proteína bruta

Introduction

Bean grains (*P. vulgaris* L.) have, from the nutritional point of view, a great importance due to the presence of characteristics that make their consumption advantageous. They contain high levels of proteins, minerals, vitamins, carbohydrates and fibers. Among these characteristics, the high protein content in the grains is remarkable, reaching values of up to 36% (Mesquita et al., 2007), and from 21.1 to 30% (Pinheiro et al., 2010). Landrace beans in particular present a variation of 17 to 32% of protein content (Pereira et al., 2011).

Beans are important sources of minerals such as calcium, iron, zinc, copper magnesium, and present particularly high potassium and phosphorus content, and low sodium content (Pereira et al., 2011). Mineral contents may vary in terms of dry matter from 0.6-2.0 g of calcium kg^{-1}; 1.2-2.2 g of magnesium kg^{-1}; 8.2-16.5 g of potassium kg^{-1}; 3.7-4.9 g of phosphorus kg^{-1}; 27.1-41.0 mg of zinc kg^{-1} and 34.8-75.8 mg of iron kg^{-1} (Ribeiro et al., 2012).

Despite the high mineral and protein content of bean grains, their bioavailability may vary according to their phytate content, known for their ability to complex with bivalent cations (calcium, iron, zinc, magnesium and copper). Phytate (myo-inositol-hexaquisphosphate) is the main form of phosphorus storage in cereal and legume grains (Gibson et al., 2010). Phytate interacts with minerals and proteins and may decrease their digestive utilization, although it has already been proven to have also beneficial effects and act as an antioxidant and anti-carcinogenic agent (Gani et al., 2012). Genetic variability of beans has already been found for phytic acid levels, ranging from 7 to 14.8 g kg^{-1}, as well as the presence of a high correlation (0.73) of phytic acid with protein (Pereira et al., 2011).

The chemical composition of plants and their grains is determined by genetics, environmental conditions and their interactions. Chemical composition affects the nutritional value and sensory properties of grains, which are also associated with genetic diversity, environmental factors and their interactions (Florez et al., 2009). Mesoamerican bean genotypes have superior mineral contents compared to those of Andean origin. Small grains (less than 16.9 g per 100 grains) are predominantly of Mesoamerican origin, and contain 90% more calcium than those of the Andean center, which are larger (more than 43.1 g per 100 grains) (Blair et al., 2010). According to Talukder et al. (2010), zinc and iron content in grains of 29 Mesoamerican bean genotypes had 16.1 and 11.3% more zinc and iron, respectively, than cultivars of Andean genotypes.

Current management systems result from the interaction of environmental, edaphic, biotic and social factors and, therefore, the grain composition has possibly been modulated by genotypes, the environment and interactions (Florez et al., 2009). In view of the cleistogamic nature and edaphic and climatic diversity on which beans have been cultivated, genetic selection and genetic drift of beans have led to the appearance of several local varieties. Different bean genotypes originated in different edaphic conditions may accumulate genetic peculiarities in relation to the absorption of nutrients and the efficiency to use them. This may be associated with the nutritional difference found in bean grain genotypes (Pinheiro et al., 2010).

The concentration of protein and minerals in grains is negatively correlated with agronomic characteristics such as number of pods per plant, number of seeds per pod and grain yield per plant (Ribeiro et al., 2008). This relationship points to the difficulty in selecting genotypes with high protein content and high yield potential. However, bean grains have shown to have positively correlated mineral contents. For example, iron content is positively correlated with magnesium, zinc, phosphorus and sulfur, and this may cause the improvement of a mineral to be associated with the simultaneous increase of other minerals (Beebe et al., 2000; Silva et al., 2012). According to Ribeiro et al. (2008), the concomitant selection for two minerals is possible, for positive linear correlations were obtained in their study.

The objective of this research was to determine the mineral, total protein and phytic acid contents in bean grains and their relationships with grain yield under the influence of different growing environments.

Materials and Methods

Twenty-six bean genotypes were used, being 22 landrace genotypes and 4 commercial genotypes; BAF 112 (IPR88-Uirapurú), BAF 115 (BRS-Valente), BAF 121 (Iapar-81) and BAF 192 (BRS-Radiante), belonging to the Active Bean Bank (Banco Ativo de Feijão BAF) of the Santa Catarina State University (UDESC) (Table 1).

Grains came from field experiments in the municipalities of Anchieta/SC, Joaçaba/SC and Lages/SC, from the 2008/2009 harvest. The municipality of Anchieta is located in the extreme west of Santa Catarina, 26°10' south latitude and 53°19' west longitude. Joaçaba is located in the mid-west of Santa Catarina, 27°10' south latitude and 51°30' west longitude. Lages is located in the southern plateau of Santa Catarina, 27°48' south latitude and 50°19' west longitude (Epagri, 2010). Data on mean daily temperature (MT), rainfall regime and distribution, and total accumulated precipitation (AP) in the municipalities where the experiments were implemented were collected (Figure 1).

The experiment had a randomized block design with 3 replicates in plots of 4 rows of 3 meters spaced 0.5 m between each other. Fifteen seeds per meter were sown; the two external lines were considered the borderlands and the useful area was delimited by the two internal lines, excluding the 0.5 meters of their ends.

Based on the recommendations for bean cultivation described by the South-Brazilian Technical Commission on Beans (2010), a base fertilization was performed along the lines prior to sowing, according to soil analysis, for a grain yield potential of 3 Mg ha^{-1}. Top-dressing fertilization was carried out twice, at the three-leaf stage and at the flowering onset (V4 and R5), using 30 kg of N (urea) per hectare in each application. The chemical characteristics of the soils (0.00-0.20 m layer) of the experimental areas before setting the experiment were: in Lages/SC, aluminum Cambisol with clayey texture, 340 g kg^{-1} of clay, 38 g kg^{-1} of O.M., pH (water) 5.8, 14.8 mg dm^{-3} of phosphorus, 168 mg dm^{-3} of potassium, 4.4 cmolc dm^{-3} of

Table 1. Identification and characteristics of the 26 bean genotypes used in the experiments conducted in Anchieta, Joaçaba and Lages.

Genotype	Center of origin	Thousand grain weight (g) [1*/]	Commercial group	Seed shape	Degree of flatness
BAF 03	Andean	312	Colors	Elliptical	Full
BAF 04	Andean	384	Colors	Elliptical	Full
BAF 07	Mesoamerican	196	Black	Oblong/Reniform long	Flat
BAF 13	Mesoamerican	238	Black	Oblong/Reniform long	Flat
BAF 23	Mesoamerican	383	Black	Oblong/Reniform long	Full
BAF 36	Mesoamerican	234	Black	Oblong/Reniform mean	Flat
BAF 42	Mesoamerican	210	Black	Spherical	Flat
BAF 44	Mesoamerican	248	Colors	Elliptical	Full
BAF 46	Andean	407	Black	Elliptical	Semi-full
BAF 47	Mesoamerican	419	Black	Elliptical	Flat
BAF 50	Mesoamerican	269	Carioca	Spherical	Flat
BAF 55	Mesoamerican	222	Black	Spherical	Flat
BAF 57	Andean	389	Black	Oblong/Reniform short	Flat
BAF 60	Mesoamerican	222	Black	Oblong/Reniform short	Flat
BAF 68	Andean	365	Colors	Elliptical	Full
BAF 75	Mesoamerican	217	Black	Oblong/Reniform long	Flat
BAF 81	Mesoamerican	205	Black	Oblong/Reniform short	Flat
BAF 84	Mesoamerican	196	Colors	Spherical	Semi-full
BAF 97	Andean	377	Black	Oblong/Reniform long	Full
BAF 102	Mesoamerican	249	Black	Oblong/Reniform short	Flat
BAF 108	Mesoamerican	249	Colors	Spherical	Full
BAF 120	Andean	496	Colors	Oblong/Reniform long	Semi-full
♦BAF 112	Mesoamerican	260	Black	Elliptical	Flat
BAF 115	Mesoamerican	252	Black	Elliptical	Semi-full
BAF 121	Mesoamerican	282	Carioca	Elliptical	Semi-full
BAF 192	Andean	566	Colors	Oblong/Reniform short	Semi-full

Source: Germplasm Bank of the CAV/UDESC. ♦ Commercial genotypes: BAF 112 (IPR88-Uirapurú), BAF 115 (BRS-Valente), BAF 121 (Iapar-81) and BAF 192 (BRS-Radiante).
*Mean value of genotypes grown at the three sites in the 2008/2009 growing season.

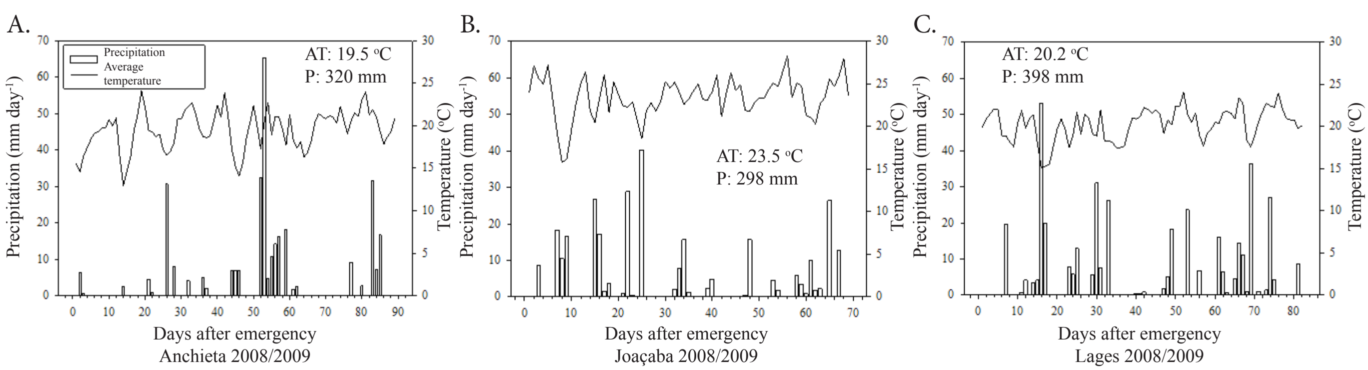

Figure 1. Rainfall and mean daily temperatures occurring between the emergence and physiological maturation of beans in (A.) Anchieta/SC, (B.) Joaçaba/SC and (C.) Lages/SC, 2008/2009 harvest.

calcium and 3.2 cmolc dm^{-3} of magnesium. In Joaçaba/SC, red clay-textured nitosol with 630 g kg^{-1} clay, M.O. 47 g kg^{-1}, pH (water) 6.1, phosphorus 19.7 mg dm^{-3}, potassium 179 mg dm^{-3}, calcium 6.7 cmolc dm^{-3} and magnesium 2.3 cmolc dm^{-3}; in Anchieta/SC, red Nitosol with clayey texture, 510 g kg^{-1} of clay, 33 g kg^{-1} of O.M., pH (water) 5.6, 9.3 mg dm^{-3} of phosphorus, 97 mg dm^{-3} of potassium, 5.5 cmolc dm^{-3} of calcium and 2.7 cmolc dm^{-3} of magnesium. Weed, disease and pest control was made with chemicals recommended for the crop. At maturity, the grains were harvested, dried, standardized to approximately 13% moisture and the grain yield was estimated in kg ha^{-1}, as the grain yield in the useful area of each plot. Afterwards, these grains were stored in a cold room until laboratory analysis at the approximate temperature of 8°C and approximate relative humidity of 40%.

The methodology for determination of nutrients and anti-nutrients was similar to that used by Pereira et al. (2011). For determination of nitrogen (N) and potassium (K), sulfur digestion was carried out. For determination of phosphorus (P), zinc (Zn) and iron (Fe) content, their total content in nitric-perchloric digestion extract was evaluated. Potassium content was assessed by obtaining a sample aliquot from sulfuric digestion, with determination of the content through emission of light in a flame photometer. Also, an aliquot from sulfur digestion was used to determine total protein content by total N content, according to the Kjeldahl method. The diluted aliquot from nitric-perchloric digestion was used to determine the concentration of iron and zinc by atomic absorption spectrophotometry. The determination of total phosphorus in bean grains was performed by the colorimetric meta-vanadate method in a sample preceded by opening through nitric-perchloric digestion. The methodology used for determination of phytic acid was similar to that described by Pereira et al. (2011), using grain samples milled in a Wiley mill and subjected to passage in anion exchange resin; at the end of the process, the supernatant was subjected to spectrophotometry under a wavelength of 500 nm.

All the studied characteristics were submitted to univariate analysis of variance obeying a linear model of fixed effects (Littel et al., 2006) for the two components, local, genotype and genotype response to the environment of the crop. The Scott-Knott test was used for comparisons between mean values of each of the analyzed variables of the different genotypes at each cultivation site, and the Tukey test was used for comparisons among sites, for each genotype. The ratio between nutrient content and grain yield was estimated through track analysis. Associations between nutrient contents were tested through simple linear correlation analysis. For all the tests performed, the probability adopted was $p \leq 0.05$.

Results and Discussion

The analysis of variance of genotypes evaluated for nutrient contents showed genotype x environment interaction for phytic acid, iron, potassium, and non-significant interaction for zinc, phosphorus and crude protein. For the grains produced in Anchieta, the genotypes showed variability for: phytate, iron, potassium and crude protein; in the grains produced in Joaçaba, the genotypes differed for phytic acid, phosphorus, iron and zinc contents; in the grains produced in Lages, the genotypes presented significant difference for phytic acid, iron, zinc, potassium and crude protein.

Total protein content of grains produced in Anchieta ranged from 238 g kg^{-1} (BAF 102) to 386 g kg^{-1} (BAF 07), averaging 292 g kg^{-1} (Table 2). This concentration was higher than that obtained in landrace bean genotypes in Spain, with values between 265-352 g kg^{-1} (Santalla et al., 2004) and in Brazil, in commercial genotypes, between 226-273 g kg^{-1} (Toledo et al., 2008). However, a greater variation of total protein in bean grains (223-363 g kg^{-1}) was observed in 21 bean lineages (Mesquita et al., 2007).

In the grains produced in Joaçaba, total protein content varied from 170 g kg^{-1} (BAF 115) to 336 g kg^{-1} (BAF 84), with a mean of 240 g kg^{-1}, but no difference was observed in protein content. In the case of grains produced in Lages, protein content varied from 241 g kg^{-1} (BAF 112) to 331 g kg^{-1} (BAF 192) with a mean of 283 g kg^{-1} (Table 2). In a study by Pereira et al. (2009) with 21 landrace bean genotypes, total protein ranged from 191 to 320 g kg^{-1}, with an average of 262 g kg^{-1}. Buratto et al. (2009) evaluated 18 bean genotypes from the black and carioca commercial groups at 3 growing sites and found genotype x environment interaction for crude protein ranging from 232 to 263 g kg^{-1} for the black commercial group genotypes and 225 to 259 g kg^{-1} for the carioca commercial group. In this work, the genotype x environment interaction for this factor was not observed, nor difference between commercial groups; however, the protein concentrations obtained by them were higher. Crude protein concentration in bean grains of Andean origin (281 g kg^{-1}) presented superiority in relation to the crude protein concentration in grains of Mesoamerican origin (267 g kg^{-1}). However, no relation was observed between thousand grain weight and crude protein content. BAF 04 and 68 stood out with crude protein content $\geq$ 300 g kg^{-1}.

It was observed that the iron content in grains produced in Anchieta ranged from 116 mg kg^{-1} (BAF 13) to 216 mg kg^{-1} (BAF 44), with a mean of 153 mg kg^{-1}. Iron content in grains

Table 2. Crude protein, iron and zinc contents of 26 bean genotypes grown in Anchieta, Joaçaba and Lages.

BAF	Crude protein (g kg^{-1})			Iron (mg kg^{-1})			Zinc (mg kg^{-1})		
	Anchieta	Joaçaba	Lages	Anchieta	Joaçaba	Lages	Anchieta	Joaçaba	Lages
03	344.2 a	182.7	300.0 a	132.2 Aa	73.6 Ab	82.8 Ab	60.0	45.6 a	48.0 a
04	361.8 a	236.1	302.2 a	149.4 Aa	96.8 Ab	66.0 Ab	50.0	44.0 a	32.0 b
07	386.1 a	222.5	295.6 a	178.8 Aa	116.8 Ab	119.4 Aa	58.0	49.2 a	46.0 a
13	266.9 b	236.1	253.7 b	215.6 Aa	216.2 Aa	114.0 Aa	48.0	41.4 a	46.0 a
23	293.4 b	275.8	290.6 a	162.4 Aa	113.4 Ab	152.2 Aa	50.0	46.6 a	50.0 a
36	304.4 b	218.4	308.9 a	153.8 Aa	102.8 Ab	136.4 Aa	48.0	49.4 a	44.0 a
42	250.7 b	238.3	273.6 b	120.4 Aa	102.0 Ab	127.2 Aa	44.0	41.0 a	38.0 b
44	251.5 b	326.5	242.7 b	116.4 Ab	88.6 Ab	75.4 Aa	44.0	47.4 a	32.0 b
46	317.7 a	214.0	315.5 a	164.8 Aa	97.4 Ab	107.6 Aa	48.0	48.0 a	50.0 a
47	317.7 a	214.0	281.2 b	132.0 Aa	71.2 Ab	91.2 Aa	44.0	45.6 a	34.0 b
50	242.7 b	266.9	251.5 b	119.6 Aa	96.4 Ab	73.0 Aa	40.0	42.8 a	28.0 b
55	284.6 b	220.6	278.0 b	211.0 Aa	102.0 Bb	115.2 Ba	52.0	46.4 a	48.0 a
57	282.4 b	183.1	286.8 a	138.2 Aa	107.6 Ab	90.2 Aa	40.0	37.6 a	36.0 b
60	295.6 b	253.7	302.2 a	155.8 Aa	82.8 Ab	98.4 Aa	50.0	35.8 a	34.0 b
68	339.7 a	269.1	297.8 a	147.0 Aa	84.2 Ab	75.8 Aa	54.0	51.6 a	48.0 a
75	250.7 b	216.2	264.7 b	165.8 Aa	106.4 Ab	88.0 Aa	52.0	44.6 a	38.0 b
81	297.8 b	328.7	282.4 b	153.8 Aa	129.6 Ab	112.6 Aa	50.0	42.0 a	46.0 a
84	269.1 b	335.3	275.8 b	175.0 Aa	127.2 Ab	97.6 Aa	36.0	34.8 b	32.0 b
97	293.4 b	172.1	315.5 a	205.4 Aa	112.8 Ab	122.0 Aa	54.0	41.2 a	42.0 a
102	238.3 b	278.0	262.5 b	125.4 Aa	86.6 Ab	98.4 Aa	44.0	43.0 a	42.0 a
108	300.0 b	262.5	289.0 a	145.8 Aa	89.8 Ab	86.6 Aa	60.0	55.0 a	44.0 a
120	295.6 b	262.5	275.8 b	137.0 Aa	96.6 Ab	88.6 Aa	48.0	46.8 a	36.0 b
♦112	271.3 b	187.5	240.5 b	136.6 Aa	99.6 Ab	97.2 Aa	38.0	42.4 a	38.0 b
115	264.7 b	169.9	258.1 b	133.0 Aa	108.2 Ab	105.2 Aa	42.0	48.6 a	44.0 a
121	284.6 b	203.0	273.6 b	139.2 Aa	100.8 Ab	85.6 Aa	40.0	42.0 a	46.0 a
192	291.2 b	271.8	330.9 a	164.2 Aa	79.2 Ab	102.6 Aa	50.0	40.6 a	40.5 a
Mean	292.2 A	240.0 C	282.6 B	153.0 A	103.5 B	100.3 B	47.8 A	44.4 B	40.9 C
MS	33.84 *	84.39 ns	17.52 *	1463.0 *	6724.0 *	843.9 *	83.79 ns	43.77 *	81.98 *
CV	13.7	26.0	9.3	29.2	29.8	24.3	17.7	12.3	18.1

Means followed by the same capital letter did not differ significantly between cultivation sites (in the lines) according to the Tukey test at 5% probability. Means followed by the same lowercase letter did not differ significantly at each culture site (in the columns) according to the Scott-Knott test at 5% probability. * Significant according to the F test at 5% probability. ♦ Commercial genotypes: BAF 112 (IPR88-Uirapurú), BAF 115 (BRS-Valente), BAF 121 (Iapar-81) and BAF 192 (BRS-Radiante).

produced in Joaçaba ranged from 71 mg kg^{-1} (BAF 47) to 130 mg kg^{-1} (BAF 81), with a mean of 103 mg kg^{-1}. The genotypes in Lages presented values between 66 (BAF 04) and 153 mg kg^{-1} (BAF 23), and the overall mean was 100 mg kg^{-1} (Table 2). According to Guzmán-Maldonado (2000) who evaluated 70 bean genotypes from two different locations in Mexico, iron content in grains ranged from 71 to 180 mg kg^{-1} at site "1" and from 64 to 280 mg kg^{-1} at site "2", evidencing that iron content dependended on the collection site and on genotypes. Iron content results in grains produced at Anchieta were superior to values described in the literature, whereas iron content of grains produced in Joaçaba and Lages were similar to works carried out with lineages that present a variation between 72-127 mg kg^{-1} (Mesquita et al., 2007), or in commercial and landrace genotypes, ranging from 60-96 mg kg^{-1} for landrace and 55-89 mg kg^{-1} in cultivated beans (Beebe et al., 2000). In a study by Pereira et al. (2011), iron content varied between 62-124 kg^{-1} in the 2005/2006 growing season and between 91-162 mg kg^{-1} in the 2006/2007 growing season.

Iron and zinc contents in grains of commercial genotypes and bean lineages are strongly affected by the genotype x environment interaction (Cichy et al., 2009; Pereira et al., 2014; Tryphone & Msolla, 2010). However, this interaction was less apparent in other studies, that is, no consensus on this theme has been reached on these minerals (Ribeiro et al., 2008; Blair et al., 2010). Iron contents in grains in the order of 59 and 57 mg kg^{-1} dry matter, respectively, have been found regardless the cultivation site (Barampama & Simard, 1993). These same authors verified that iron and zinc contents were significantly influenced by the cultivar and the cultivation site, while Ribeiro et al. (2008) found no significant effect. According to Ribeiro et al. (2012), iron concentration was higher in the bean seed tegument when compared to the embryo (embryo + cotyledons) and an inverse relationship between accumulation of iron in the embryo and the tegument of the bean grain was observed. In addition, according to Beebe et al. (2000) and Ribeiro et al. (2012), the concentration of iron in the tegument of bean grains was higher in the black commercial group due to the higher concentration of tannin, a substance that has the capacity to complex with iron. In this study, genotypes of the black commercial group had a higher average iron concentration (133 mg kg^{-1}) than the other genotypes (106 mg kg^{-1}). A higher concentration of iron in genotypes of Mesoamerican origin (126 mg kg^{-1}) compared to those of Andean origin (113 mg kg^{-1}) was also observed. However, no correlation was found between grain size and iron.

Zinc content did not present genotype x environment interaction, indicating that the environment has less influence over the accumulation of this element in the grain. This can be convenient for the selection of genotypes. Zinc content in grains at Anchieta ranged from 36 (BAF 84) to 60 mg kg^{-1} (BAF 03), with a mean of 48 mg kg^{-1}. In genotypes produced in Joaçaba, zinc content varied from 35 (BAF 84) to 55 mg kg^{-1} (BAF 108), with a mean value of 44 mg kg^{-1}. The genotypes in Lages presented zinc contents ranging from 28 (BAF 50) to 50 mg kg^{-1} (BAF 23) with a mean of 40 mg kg^{-1} (Table 2). The values found in samples from Anchieta were higher than those reported by Beebe et al. (2000) and Ribeiro et al. (2012), who observed a variation from 29 to 43 mg kg^{-1} for wild genotypes and from 35- to 54 mg kg^{-1} for commercial genotypes. However, they were similar to those found by Pereira et al. (2011) when evaluating the zinc content in landrace beans grown in two harvests. The characterization of landrace genotypes is pointed out as an important factor by other authors, suggesting that iron and zinc content of bean grains can be increased in landrace genotypes. These studies report an increase in iron content between 60 and 80% and an increase in zinc of 50% (Beebe et al., 2000; Pereira et al., 2014). The concentration of zinc in beans of Andean origin (46 mg kg^{-1}) was higher than that in grains of Mesoamerican origin (44 mg kg^{-1}). It is noteworthy that BAF 03 and 68 had zinc content higher than 51 mg kg^{-1}. However, no relation was observed between grain size and zinc content. Pinheiro et al. (2010) also found no relationship between zinc concentration and seed size.

Phosphorus content varied from 3 (BAF 50) to 5 g kg^{-1} (BAF 04) in Anchieta, from 3 (BAF 55) to 6 g kg^{-1} (BAF 04) in Joaçaba, and and from 3 (BAF 112) to 5 g kg^{-1} (BAF 46) in Lages. The average phosphorus contents in Anchieta, Joaçaba and Lages were approximately 4 g kg^{-1} (Table 3). These contents were similar to those found in commercial genotypes, 4-5 g kg^{-1} (Pereira et al., 2011).

Potassium contents in the grains ranged from 14 (BAF 97) to 22 g kg^{-1} (BAF 07) in Anchieta, 15 (BAF 102) to 20 g kg^{-1} (BAF 42) in Joaçaba, and 15 (BAF 115) to 20 g kg^{-1} (BAF 46) in Lages. The average potassium contents contents were 16, 17 and 17 g kg^{-1} in Anchieta, Joaçaba and Lages, respectively (Table 3). According to Ribeiro et al. (2012), the highest potassium concentration in grains happens in embryo + cotyledons (76 to 90%).

Phytic acid content in grains produced in Anchieta ranged from 3 (BAF 84) to 14 g kg^{-1} (BAF 07), with an average value of 10 g kg^{-1}. In Joaçaba, the phytic acid content varied from 7 (BAF 112) to 14 g kg^{-1} (BAF 75), with a overall average of 10 g kg^{-1}. In Lages, the phytic acid content varied from 9 (BAF 112) to 14 g kg^{-1} (BAF 36) with a mean of 10 g kg^{-1} (Table 3). The phytic acid contents found here were higher than those found by Doria et al. (2012), who observed a variation of 4 to 8 g kg^{-1}. According to Ariza-Nieto et al. (2007), Mesoamerican genotypes have higher concentrations of phytate than those of Andean origin. In contrast with the assertion of these authors, no relationship was found here between the phytic acid contents and the center of origin, neither with grain size.

As regards grain yield (Table 4; compiled from Zilio et al., 2011), it was observed that BAFs (genotypes) presented higher grain yield in Joaçaba, with highlight to BAFs 42; 57; 102; 112 and 121, which presented average grain yield above 4,000 kg ha^{-1} in Joaçaba. In Anchieta, the BAF 55 stood out, and in Lages, the BAF 13 stood out with grain yield above 3,000 kg ha^{-1}. The genotypes BAF 13; BAF 50; BAF 55; BAF 81 and BAF 121 presented grain yields above 2,000 kg ha^{-1} in the three growing sites.

The track analysis (Table 5) showed that grain yield had a negative correlation with phytic acid (-0.14*), zinc (-0.33*), phosphorus (-0.39*), potassium (-0.38*) and crude protein (-0.50*) contents, indicating that genotypes that present high grain yield tended to have lower concentrations of nutrients.

Table 3. Phosphorus, phytic acid and potassium contents of grains of 26 bean genotypes grown in Anchieta, Joaçaba and Lages.

BAF	Phosphorus (g kg^{-1})			Phytic acid (g kg^{-1})			Potassium (g kg^{-1})		
	Anchieta	Joaçaba	Lages	Anchieta	Joaçaba	Lages	Anchieta	Joaçaba	Lages
03	4.3	4.8	4.1	7.1 Ab	9.0 Aa	11.6 Aa	16.6 b	18.3	17.2 a
04	4.8	5.2	3.5	12.2 Aa	13.4 Aa	11.5 Ab	15.5 b	17.9	16.5 a
07	4.6	4.1	3.5	13.9 Aa	11.3 Aa	12.6 Aa	22.4 a	17.9	18.7 a
13	3.2	3.6	3.2	5.3 Ab	10.1 Aa	10.3 Ab	14.9 b	16.9	17.3 a
23	3.9	4.1	4.1	4.3 Bb	10.9 Aa	9.8 ABb	16.2 b	16.9	17.3 a
36	3.8	3.8	4.3	11.2 Aa	12.5 Aa	13.5 Aa	15.7 b	16.9	17.6 a
42	3.6	3.3	3.2	11.5 Aa	10.7 Aa	11.2 Aa	15.1 b	19.4	16.9 a
44	3.5	3.9	3.5	9.5 Aa	10.6 Aa	9.2 Aa	16.8 b	16.9	16.6 a
46	3.8	3.8	4.8	10.0 Aa	13.1 Aa	13.3 Aa	17.5 b	19.4	19.9 a
47	4.5	4.1	4.0	9.9 Aa	11.7 Aa	9.7 Ab	15.7 b	17.5	16.5 a
50	3.0	3.5	2.9	9.6 Aa	11.4 Aa	11.7 Ab	15.0 b	15.3	15.7 a
55	3.2	2.6	4.0	9.8 Aa	10.2 Aa	12.5 Aa	15.1 b	16.2	17.2 a
57	3.5	3.3	3.5	8.9 Aa	11.8 Aa	11.8 Aa	15.7 b	17.6	16.1 a
60	4.0	3.1	3.9	8.3 Aa	10.4 Aa	11.0 Aa	16.6 b	18.2	17.3 a
68	4.1	4.4	4.2	13.1 Aa	11.7 Aa	12.5 Aa	16.8 b	17.2	17.2 a
75	3.7	3.6	3.4	12.4 Aa	14.0 Aa	10.7 Ab	16.0 b	19.1	17.5 a
81	4.1	3.3	4.3	11.6 Aa	11.3 Aa	10.3 Ab	15.7 b	15.7	16.4 a
84	3.2	4.3	3.7	2.9 Bb	11.9 Aa	10.8 Ab	15.5 b	16.1	16.5 a
97	3.8	3.8	3.9	9.8 Aa	8.0 Aa	12.4 Aa	14.4 b	17.2	17.3 a
102	3.1	3.6	2.8	11.1 Aa	12.1 Aa	11.4 Aa	15.3 b	14.9	16.5 a
108	3.9	3.6	3.8	10.1 Aa	10.8 Aa	10.3 Ab	16.5 b	16.2	18.0 a
120	3.9	4.0	2.9	10.5 Aa	12.0 Aa	10.6 Ab	16.4 b	15.1	16.9 a
♦112	3.2	3.3	2.7	9.2 Aa	7.1 Ab	9.0 Ab	16.0 b	16.8	15.7 a
115	3.9	3.3	4.0	10.0 Aa	11.4 Aa	10.4 Ab	16.4 b	16.8	15.4 b
121	3.1	3.0	3.4	9.4 Aa	8.3 Aa	10.4 Ab	15.1 b	16.0	17.2 a
192	4.1	4.0	3.8	12.3 Aa	11.0 Aa	11.5 Aa	17.5 b	16.8	17.9 a
Mean	3.8 B	3.8 B	4.2 A	9.8 A	10.1A	10.1 A	16.2 B	17.0 A	17.0 A
MS	0.49 ^{ns}	0.65 *	16.65 ^{ns}	0.13 *	0.05 *	0.03 *	4.46 *	2.98 ^{ns}	1.83 *
CV	18.5	18.5	18.8	28.4	17.1	11.6	10.9	9.1	6.8

Means followed by the same capital letter did not differ significantly between cultivation sites (in the lines) according to the Tukey test at 5% probability. Means followed by the same lowercase letter did not differ significantly at each culture site (in the columns) according to the Scott-Knott test at 5% probability. * Significant according to the F test at 5% probability. ♦ Commercial genotypes: BAF 112 (IPR88-Uirapurú), BAF 115 (BRS-Valente), BAF 121 (Iapar-81) and BAF 192 (BRS-Radiante).

Table 4. Grain yield of the 26 bean genotypes in the three growing sites, 2008/2009 harvest.

Genotype	Yield (kg ha^{-1})		
	Anchieta	Joaçaba	Lages
03	1099.1	2616.7	1220.1
04	841.5	2010.9	1656.1
07	493.3	2776.0	1937.7
13	2351.4	3768.2	3064.6
23	925.5	2788.2	570.6
36	2203.5	3327.0	1605.0
42	1986.8	4451.9	2470.0
44	1514.8	2282.3	1957.1
46	1909.3	3723.8	1540.6
47	2122.4	2661.1	1107.2
50	2298.6	3665.4	2646.1
55	3536.6	3766.5	1806.0
57	1875.9	5096.1	491.8
60	1544.7	3941.4	1713.8
68	1183.6	2761.0	750.1
75	2704.3	3955.9	1519.1
81	2117.2	3693.0	2329.9
84	2351.4	3948.7	1430.3
97	1181.9	3103.7	1058.6
102	1823.0	5018.1	2672.6
108	1106.1	2750.5	1887.7
120	804.9	2822.5	981.5
♦112	2306.8	4385.8	1931.1
115	2601.5	3856.0	1871.0
121	2080.2	4019.4	2524.1
192	1398.3	3246.8	705.7

♦Commercial genotypes: BAF 112 (IPR88-Uirapurú), BAF 115 (BRS-Valente), BAF 121 (Iapar-81) and BAF 192 (BRS-Radiante). Source: Zilio et al. (2011).

Iron content had a positive but low (0.03^{ns}) correlation coefficient, indicating a lack of association with grain yield.

In Joaçaba, grain yield presented a negative correlation with zinc (-0.45*), phosphorus (-0.58*) and crude protein (-0.11^{ns}). Iron content presented a positive correlation (0.14*) with grain yield, indicating that higher iron content was found in more productive genotypes. The correlation between phytate (-0.09^{ns}), and potassium (-0.07^{ns}) evidenced a lack of association with grain yield. In the environment of Lages, only phytic acid (-0.11^{ns}) and crude protein (-0.42*) contents were associated with grain yield. Iron, zinc, phosphorus and potassium contents showed very low correlation coefficients with grain yield, evidencing absence of relation.

High grain yields are associated with lower nutrients contents, especially nitrogen (crude protein), but the contribution of these characteristics to grain yield is differentiated and dependent on the location of the crop (Figure 2).

According to Ribeiro et al. (2008), negative linear correlation coefficients were obtained between grain yield and boron (-0.50*), copper (0.53*) and zinc (-0.53*) contents. Leleji et al. (1972) found that bean lineages with higher grain yield had lower nitrogen content and, consequently, lower crude protein in grains.

Regarding the direct effects on grain yield in Anchieta, zinc (-0.20*), phosphorus (-0.11^{ns}), potassium (-0.38*) and crude protein (-0.29*) contents presented negative correlation coefficients. For iron contents, a positive correlation (0.21*) was observed, indicating that the higher the grain yield, the higher is the iron concentration in bean grains. In the environment of Joaçaba, phosphorus content (-0.54*) presented a correlation with grain yield, followed by zinc (-0.36*), iron (-0.12^{ns}) and potassium (-0.11^{ns}) contents. Phytic acid (0.06^{ns}) and crude protein (-0.04^{ns}) contents showed an extremely low direct

Table 5. Phenotypic estimates of the direct and indirect effects of grain yield and nutrient content of the 26 bean genotypes grown at three different locations in Santa Catarina.

Characteristics	Correlation estimate		
	Anchieta	Joaçaba	Lages
Phytate			
Direct effect on grain yield	0.09	0.06	0.09
Indirect effect via iron	-0.03	0.002	0.02
Indirect effect via zinc	-0.03	-0.05	0.02
Indirect effect via phosphorus	-0.04	-0.09	0.01
Indirect effect via potassium	-0.06	-0.007	0.008
Indirect effect via crude protein	-0.07	-0.0006	-0.26*
Total	-0.14*	-0.08	-0.11
Iron			
Direct effect on grain yield	0.21	-0.12	0.11
Indirect effect via phytate	-0.01	-0.001	0.015*
Indirect effect via zinc	-0.11	0.02	0.032*
Indirect effect via phosphorus	-0.03	0.24*	-0.008
Indirect effect via potassium	-0.05	0.008	0.003
Indirect effect via crude protein	-0.01	0.0004	-0.1
Total	0.03	0.14*	0.04
Zinc			
Direct effect on grain yield	-0.2	-0.37*	0.07
Indirect effect via phytate	0.01	0.008	0.02
Indirect effect via iron	0.1	0.006	0.05
Indirect effect via phosphorus	-0.06	-0.13*	-0.01
Indirect effect via potassium	-0.07	0.017*	0.007
Indirect effect via crude protein	-0.11	0.003	-0.14*
Total	-0.33*	-0.46*	-0.007
Phosphorus			
Direct effect on grain yield	-0.11	-0.54*	-0.09
Indirect effect via phytate	0.03	0.009	-0.01
Indirect effect via iron	0.005	0.05	0.009
Indirect effect via zinc	-0.11	-0.09	0.008
Indirect effect via potassium	-0.06	-0.008	-0.002
Indirect effect via crude protein	-0.15*	-0.009	0.05
Total	-0.4	-0.58	-0.04
Potassium			
Direct effect on grain yield	-0.23*	-0.11	0.02
Indirect effect via phytate	0.02	0.004	0.03
Indirect effect via iron	0.05	0.008	0.01
Indirect effect via zinc	-0.06	0.06	0.02
Indirect effect via phosphorus	-0.03	-0.04	0.008
Indirect effect via crude protein	-0.13*	0.007	-0.19*
Total	-0.38*	-0.07	-0.1
Crude protein			
Direct effect on grain yield	-0.29*	-0.04	-0.52*
Indirect effect via phytate	0.02	0.0009	0.04
Indirect effect via iron	0.01	0.001	0.02
Indirect effect via zinc	-0.08	0.03	0.02
Indirect effect via phosphorus	-0.05	-0.12*	0.008
Indirect effect via crude protein	-0.1	0.02	0.008
Total	-0.5	-0.11*	-0.42*

* Significant according to the t-test at 5% probability.

correlation, indicating an absence of direct association between these and grain yield in Joaçaba. In Lages, only crude protein contents presented a negative correlation and a significant effect on grain yield (-0.52*). Iron content presented a positive correlation coefficient (0.10^{ns}), similar to the results found in Anchieta.

The variation in the magnitude of the correlations obtained in the different locations is indicative of the need for evaluating the genotypes in different environments, to know how the environment influences the nutrient composition of grains and its correlation with grains yield.

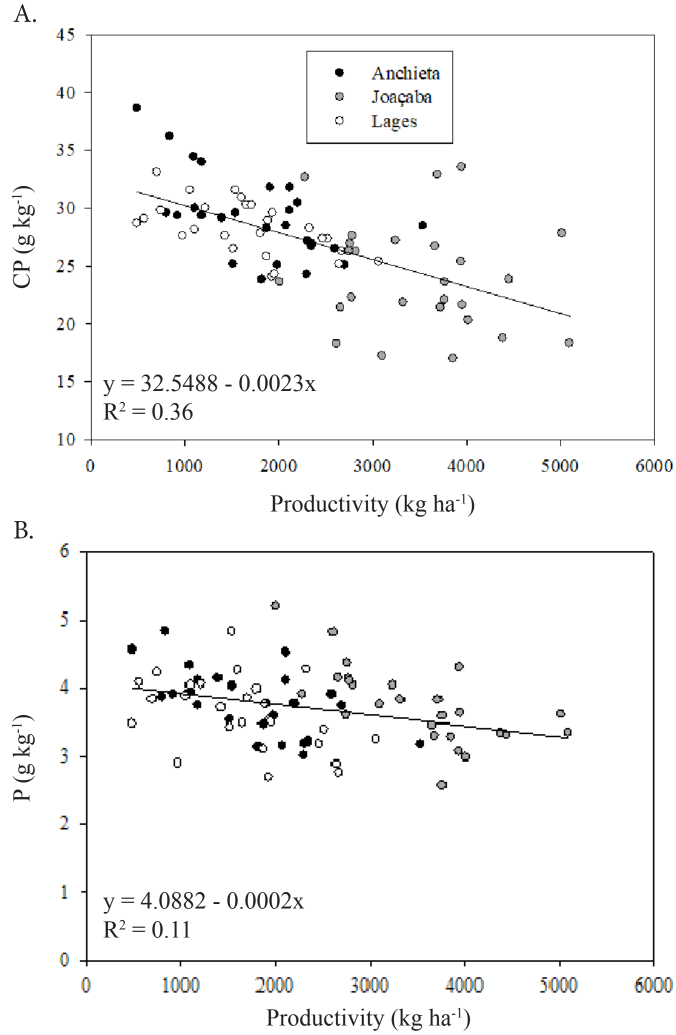

Figure 2. Relationship of crude protein (A) and phosphorus (B) contents with grain yield of the 26 bean genotypes grown in Anchieta, Joaçaba and Lages.

Based on the indirect effect of the nutrients on the basic grain yield variable, the correlation coefficients were low in the three sites, indicating an absence of correlation. Therefore, in general, the indirect selection of nutrient contents and grain yield does not seem to be a good strategy for genetic progress because most of the correlation coefficients were very low. In terms of direct effects on grain yield, nitrogen is the most affected nutrient by high grain yield at the three growing sites. Grains produced in Anchieta and Joaçaba presented reduced zinc, phosphorus and potassium contents when grain yield was increased. This shows the difficulties in identifying genotypes with high nutrient levels and grain yield, considering that the association between these characteristics is negative, that is, the increase of one parameter results in the decrease of the other.

The concomitant choice for two minerals was possible because positive and significant linear correlation coefficients were obtained, but there were differences in nutrients and in the magnitude of the correlations depending on cultivation site. In Anchieta, grains of the genotypes showed a positive and significant correlation between zinc and iron (0.49*), zinc and phosphorus (0.52*), zinc and potassium (0.32*), phytic acid and phosphorus (0.35*), and phytic acid and potassium (0.27^{ns}).

In Lages, there was a correlation between zinc and potassium (0.32*), zinc and iron (0.48*), phytic acid and zinc (0.28*) and phytic acid and potassium (0.37*). However, genotypes grown in Joaçaba showed no significant correlation between minerals. Therefore, the results found provide evidence of the influence of the environment on nutrient contents and their inter-relationships. This allows us to say that nutrient content assessments in bean grains must be carried out at different sites in order to determine the capacity and variation of nutrient accumulation in the grains of a genotype and that the increase in one nutrient may lead to the increase of another depending on the place of cultivation. Such observations are important for genotype selection. Pinheiro et al. (2010) evaluated 155 bean genotypes from different centers of origin and found an association between Zn-Fe, Zn-Cu, Cu-P, and Ca-Mn. Positive correlations between several minerals have also been observed in genotypes of Andean and Mesoamerican bean genotypes (Beebe et al., 2000; Cichy et al., 2009; Pinheiro et al., 2010; Talukder et al., 2010), highlighting that the selection to increase the content of a mineral will result in the increment of another.

Phytic acid is considered an anti-nutrient because of the absence of phytase enzyme in the intestinal tract of humans and monogastric organisms. The ingestion of food containing phytic acid causes the availability of nutrients to be drastically reduced when consuming grains containing high levels of phytic acid (Gani et al., 2012). Thus, the positive correlations found between minerals and phytate are not favorable because the nutrients are complexed with phytic acid, which makes it an important anti-nutrient in grains, particularly in legumes such as beans, taking into account that phytic acid is located in protein bodies present inside the cotyledons (Gibson et al., 2010).

Conclusions

Landrace genotypes showed a great diversity in nutrient content depending on the culture site. Iron, potassium and phytic acid contents varied according to the culture site, indicating that the environment influences the content of these nutrients in the evaluated genotypes. However, in the case of zinc, phosphorus and crude protein content, the effect of genotype is preponderant.

The concomitant choice for two minerals such as zinc-iron and zinc-potassium is possible in environments of Lages and Anchieta.

High grain yields are related with lower phosphorus and nitrogen (crude protein) content in the grains.

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