

# GENOTYPIC VARIABILITY IN SEED ACCUMULATION OF FOLIAR-APPLIED MOLYBDENUM TO COMMON BEAN<sup>(1)</sup>

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## SUMMARY

The genotypic variability in molybdenum (Mo) accumulation in common bean seeds has been demonstrated in cases in which soil is the main Mo source, but this variability is yet unknown when Mo is foliar-applied. Therefore, seed Mo concentrations (SMoCc) and seed Mo contents (SMoCt) of 12 genotypes were determined in four experiments in the Zona da Mata, Minas Gerais, Brazil, in which plants were sprayed with 600 g ha<sup>-1</sup> Mo. For comparison, two additional experiments without external Mo were conducted. Without Mo application, the average SMoCc was undetectable or 2.83 µg g<sup>-1</sup>, without significant differences among genotypes. On average, with Mo applications, SMoCc ranged from 14.7 to 25.0 µg g<sup>-1</sup> and SMoCt, from 3.94 to 6.84 µg. 'Majestoso' was among the genotypes with the highest SMoCc in the four experiments. However, the large-seeded 'Jalo MG-65' and 'Carnaval' generally had higher SMoCt than the small-seeded 'Majestoso'. 'Ouro Negro' and especially 'Valente' were among the genotypes with the lowest SMoCc and SMoCt. The values of these variables were 61 and 90 %, respectively, higher for 'Majestoso' than those for 'Valente'. Our results suggest that common bean genotypes differ in their capacity to accumulate foliar-applied Mo in the seeds. Mo-rich seeds of large-seeded genotypes or of small-seeded of small-seeded genotypes with good capacity to accumulate Mo in seeds can be produced with relatively less Mo fertilizer.

**Index terms:** *Phaseolus vulgaris*, seed molybdenum content, foliar fertilization, growth habit.

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## RESUMO: VARIABILIDADE GENOTÍPICA EM FEIJÃO-COMUM EM ACUMULAR NA SEMENTE O MOLIBDÊNIO APLICADO NA FOLHAGEM

A variabilidade genotípica em feijão-comum em acumular molibdênio (Mo) nas sementes foi demonstrada quando o solo é a principal fonte de Mo; entretanto, essa variabilidade ainda é desconhecida quando o Mo é aplicado na folhagem. Portanto, determinaram-se teores de Mo da semente (TMoS) e conteúdos de Mo da semente (CMoS) de 12 genótipos em quatro experimentos conduzidos na Zona da Mata, Minas Gerais, Brasil, nos quais pulverizaram-se as plantas com 600 g ha<sup>-1</sup> de Mo. Para comparação, conduziram-se mais dois experimentos sem aplicação de Mo. Nesse caso, não foi detectado Mo na semente ou o TMoS médio foi 2,83 µg g<sup>-1</sup>, sem diferença significativa entre genótipos. Em média, com aplicação de Mo, os TMoS variaram de 14,7 a 25,0 µg g<sup>-1</sup> e os CMoS, de 3,94 a 6,84 µg. 'Majestoso' ficou entre os genótipos com os mais altos TMoS nos quatro experimentos. No entanto, as sementes grandes de 'Jalo MG-65' e 'Carnaval' geralmente apresentaram mais altos CMoS que as sementes pequenas da 'Majestoso'. 'Ouro Negro' e, sobretudo, 'Valente' ficaram entre os genótipos com os mais baixos TMoS e CMoS. Os valores dessas variáveis foram 61 e 90 %, respectivamente, mais altos na 'Majestoso' que na 'Valente'. Os resultados sugerem que genótipos de feijão-comum diferem na capacidade de acumular nas sementes o Mo aplicado na folhagem. Sementes enriquecidas com Mo de genótipos de sementes grandes ou de genótipos de sementes pequenas bons acumuladores de Mo na semente podem ser produzidas com dose relativamente menor de fertilizante molibdic.

*Termos de indexação:* Phaseolus vulgaris, conteúdo de molibdênio da semente, adubação foliar, hábito de crescimento.

## INTRODUCTION

Molybdenum (Mo) is a constituent of three enzymes related to the nitrogen (N) nutrition of plants: nitrogenase, nitrate reductase and xanthine dehydrogenase. These enzymes act, respectively, in the acquisition of N by means of biological N fixation, in N utilization absorbed as nitrate, and in the degradation of purine (Mendel, 2011). The N concentration in Mo-deficient common bean plants may therefore be low, due to the reduced activity of these enzymes. Xanthine dehydrogenase is also involved in plant resistance to disease and drought and leaf senescence. Two other plant enzymes contain Mo: sulfide oxidase and aldehyde oxidase. The latter is involved in the synthesis of abscisic acid, which plays an important role in plant growth and development and in the adaptive response to various stress environments (Mendel, 2011). Sulfide oxidase is involved in cell detoxification of excess sulfide (Mendel, 2011) and in the degradation of sulfur-containing amino acids (Kaiser et al., 2005).

In the region of Zona da Mata of Minas Gerais, Mo applied to common bean leaves is the most commonly used method to correct soil Mo deficiency. Treating seeds with molybdenum fertilizer is also effective, but can be toxic to rhizobia (Campo et al., 2009). The plant use efficiency of Mo applied to foliage is higher than that of Mo applied to the soil along with other fertilizers (Abreu et al., 2007). Therefore, the Mo rate required for foliar application is lower than for soil application. Foliar Mo application also allows an even field distribution at low cost, because the Mo fertilizer can be added to the pesticide solution (Silva et al., 2003). In the region of Zona da Mata,

foliar Mo fertilization should be applied between 14 and 28 d after emergence (DAE) (Berger et al., 1996) at a rate between 70 and 100 g ha<sup>-1</sup> Mo (Berger et al., 1996; Amame et al., 1999; Pessoa et al., 2001). In this region, Mo fertilization can increase common bean yields up to three times (Pessoa et al., 2001), with no need for N topdressing (Vieira et al., 1992, Berger et al., 1996; Amame et al., 1999).

The plant requirement for Mo is one of the lowest, compared to the other micronutrients. Therefore, large seeds as those of common bean (*Phaseolus vulgaris* L.) and soybean [*Glycine max* (L.) Merr.] can supply a plant with Mo when the soil is Mo-deficient. Mo-rich seeds of common bean and soybean can be produced in Mo-poor soils. To enrich these seeds with Mo in these soils, the plants are sprayed with Mo rates between 240 and 1,600 g ha<sup>-1</sup> (Vieira et al., 2005; Kubota et al., 2008; Campo et al., 2009; Vieira et al., 2011a,b; Pacheco et al., 2012; Almeida et al., 2013). Molybdenum applied to the foliage can be transferred within 24 h to other plant parts (Williams et al., 2004). This rapid Mo transport in the plant probably also occurs when Mo-rich seeds are used. Therefore, the Mo contained in the seed becomes available to the plant quicker than that applied to the foliage. This prompt plant availability when Mo-rich seeds are used can benefit nitrogenase activity in the early vegetative stage of common bean (Almeida et al., 2013). Small farmers can benefit most from the use of these seeds, because they usually have no access to Mo fertilizer or are unaware of the advantages Mo fertilization can have on common bean.

In soybean, spraying plants with a total rate of 800 g ha<sup>-1</sup> Mo increased the seed Mo content by 3,000 % (Campo et al., 2009). In regions with Mo-poor soils, as

in Zona da Mata, plants grown from common bean seeds containing  $3.6 \pm 0.7 \mu\text{g Mo}$  per seed or more (high Mo content) did not respond to Mo spraying, unlike seeds with low Mo content (Vieira et al., 2011b). Seeds with high Mo content were derived from plants sprayed with  $1,000 \text{ g ha}^{-1} \text{ Mo}$  and those with low Mo content ( $0.007 \pm 0.001$  or  $0.248 \pm 0.057 \mu\text{g Mo}$  per seed) from plants sprayed with water or  $90 \text{ g ha}^{-1} \text{ Mo}$ , respectively (Vieira et al., 2010). Split Mo applications can induce higher seed Mo contents than a single application (Vieira et al., 2005).

The price of Mo increased greatly in recent years (Guang-Yi et al., 2012). One way to use Mo fertilizer efficiently to increase Mo content in seed is to know the genotypic variability in the capacity to accumulate foliar-applied Mo in the seeds. When the soil is the main Mo source, the common bean genotypes differ in the capacity to accumulate Mo in seeds (Franco & Munns, 1981; Brodrick & Giller, 1991b; Brodrick et al., 1995). However, the genotypic variability in the capacity to accumulate Mo in seeds has not been studied so far for foliar-applied Mo. Our objective was to evaluate genotypes from four Brazilian common bean breeding programs for their capacity to accumulate foliar-applied Mo in the seeds.

## MATERIAL AND METHODS

Four field experiments were conducted in which plants of 12 common bean genotypes were sprayed with  $600 \text{ g ha}^{-1} \text{ Mo}$ . Locations and soils of common bean fields in the Zona da Mata, State of Minas Gerais, Brazil, were chosen where common bean had responded positively to foliar-applied Mo in previous years. In Oratórios, two experiments were installed in 2008 (April 10 and July 11) and one in 2010 (March 18). In Coimbra, one experiment was installed on March 29, 2010. In 2010, adjacent to the two experiments with Mo application (Coimbra and Oratórios), an experiment without external Mo application was carried out for comparison.

In Oratórios, the experiments were carried out at an experimental station ( $20^{\circ} 24' 12'' \text{ S}$ ,  $42^{\circ} 49' 08'' \text{ W}$ ; 478 m) of the Agricultural Research Institute of Minas Gerais (Epamig); in Coimbra, in an area ( $20^{\circ} 49' 44'' \text{ S}$ ,  $42^{\circ} 45' 47'' \text{ W}$ ; 716 m) of the Federal University of Viçosa (UFV). The soils of these areas are classified as Ultisol (Embrapa, 2006), with  $560 \text{ g kg}^{-1}$  clay,  $190 \text{ g kg}^{-1}$  silt, and  $250 \text{ g kg}^{-1}$  sand in Coimbra, and 270, 150 and  $580 \text{ g kg}^{-1}$ , respectively, in Oratórios. Other soil properties are shown in table 1. The soils were prepared by disc plowing followed by three diskings before planting.

Eleven common bean cultivars and line VC-8 were used (Table 2). These genotypes were derived from four common bean breeding programs in Brazil. The number of genotypes of each plant Type (III, II or I)

represents approximately the proportion of use of each of these types by Brazilian farmers. Type III plants have a semiprostrate to prostrate indeterminate growth habit; Type II upright indeterminate growth habit; and Type I plants have a determinate growth habit. The cultivars Jalo MG 65 and Carnaval MG have large seeds (Andean origin) and the other genotypes small seeds (Mesoamerican origin). The seeds of the 12 genotypes were obtained from the UFV. These seeds were sown together for seed multiplication, without external Mo application, in an experimental area of the UFV in Coimbra.

A randomized block design with five (April 2008) or four replications (other experiments) was used. The plots consisted of one (2008) or three (2010) 2-m rows, spaced 0.5 m apart, with a density of 15 seeds per meter. In 2010, the outer rows were the borders. In 2008, the genotypes of neighboring plots were the lateral borders, and cultivar Ouro Vermelho was planted as the external border of the experiments. Therefore, an area of  $1 \text{ m}^2$  was evaluated in both years.

In 2008, Mo (in  $\text{g ha}^{-1}$ ) as sodium molybdate was split as follows: 300 in the V4 growth stage (third trifoliate leaf) and 300 in the R6 stage (flowering). In that year, the early maturing cultivar Carnaval was sown on the same day as the others. On the days of Mo spraying, the plants of Carnaval were in the R5 stage (pre-flowering) and R7 stage (early pod formation), respectively. In 2010, Mo (in  $\text{g ha}^{-1}$ ) was split in: 100 in V4, 250 in R5 and 250 in R7. At that time, cultivar Carnaval was sown 7 d after the others so that Mo was sprayed in the three scheduled growth stages. In both experiments in 2010, in which no Mo was applied, the genotypes were sown on the same day. A  $\text{CO}_2$  sprayer equipped with two XR 11002 cone nozzles, spaced 0.5 m apart, with a flow rate of  $225 \text{ L ha}^{-1}$  at a pressure of 207 kPa was used. Dodecylbenzene sulfonic acid spreader-sticker at  $20 \text{ g L}^{-1}$  was added to the Mo solution.

In Coimbra, a basal fertilization of  $24 \text{ kg ha}^{-1} \text{ N}$ ,  $36 \text{ kg ha}^{-1} \text{ P}$ , and  $39 \text{ kg ha}^{-1} \text{ K}$  was applied in the sowing furrow; in Oratórios,  $14 \text{ kg ha}^{-1} \text{ N}$ ,  $21 \text{ kg ha}^{-1} \text{ P}$ , and  $23 \text{ kg ha}^{-1} \text{ K}$ . Both fertilizations were based on recommendation of Barbosa & Gonzaga (2012) for target yields above  $2500 \text{ kg ha}^{-1}$ . Rhizobium inoculant was not used. In the season prior to the experiments, the common bean plants grown in the plots had produced many nodules, indicating the presence of native rhizobia strains in these soils. Around 20 DAE,  $150 \text{ kg ha}^{-1}$  urea was applied along the rows, as recommended by Barbosa & Gonzaga (2012), for target yields between 2500 and  $3500 \text{ kg ha}^{-1}$ . Weeds were controlled with fomesafen ( $150 \text{ g ha}^{-1}$ ) and fluzafop-p-butyl ( $200 \text{ g ha}^{-1}$ ) and, where necessary, also with surface hoeing. Pests were controlled with methamidophos ( $500 \text{ mL ha}^{-1}$ ) or chlorpyrifos ( $400 \text{ mL ha}^{-1}$ ). Leaf diseases were preventively controlled with epoxiconazole ( $12.5 \text{ mL ha}^{-1}$ ) and azoxystrobin ( $50 \text{ g ha}^{-1}$ ), applied alternately. At sowing, and then every 3 d until

**Table 1. Chemical characteristics of soils in the 0-20 cm layer in the experimental areas prior to the experiments**

Soil property <sup>(1)</sup>	Oratórios		Coimbra	
	April 2008 <sup>(2)</sup>	July 2008	March 2010	March 2010
pH (H <sub>2</sub> O) (1:2.5)	6.4	6.4	5.8	4.7
P, mg dm <sup>-3</sup>	32	73	31	11
K, mg dm <sup>-3</sup>	120	132	112	84
Al, cmol <sub>c</sub> dm <sup>-3</sup>	0.0	0.0	0.0	0.5
Ca, cmol <sub>c</sub> dm <sup>-3</sup>	2.4	3.5	2.9	1.4
Mg, cmol <sub>c</sub> dm <sup>-3</sup>	1.1	1.2	0.9	0.5
Base saturation, %	79	79	59	31
H+Al, cmol <sub>c</sub> dm <sup>-3</sup>	0.99	1.32	2.81	4.79
CEC (T), cmol <sub>c</sub> dm <sup>-3</sup>	4.80	6.36	6.90	6.90
Organic matter, dag kg <sup>-1</sup>	1.1	2.1	2.5	3.0

<sup>(1)</sup> P and K extracted by Mehlich-1 solution. Al, Mg, and Ca extracted by HCl 1 mol L<sup>-1</sup>. Base saturation = [(K + Ca + Mg)/T] x 100, in which T = K + Ca + Mg + (H+Al); H+Al extracted by a solution of calcium acetate 0.5 mol L<sup>-1</sup>, pH 7.0. <sup>(2)</sup> Location and month and year of the installation of the experiment.

**Table 2. Seed and plant characteristics of common bean genotypes and institutions of origin**

Genotype	Commercial class <sup>(1)</sup>	Seed size	Plant type <sup>(2)</sup>	Origin <sup>(3)</sup>
Ouro Vermelho	red	small	III	UFV
Pérola	carioca	small	III	Embrapa
BRSMG Majestoso	carioca	small	III	UFLA
VC-8	carioca	small	III	UFV
Ouro Negro	black	small	III	Embrapa
BRSMG Madrepérola	carioca	small	III	UFV
BRSMG Talismã	carioca	small	III	UFLA
BRSMG Pioneiro	carioca	small	II	UFV
Jalo MG-65	jalo	large	III	Epamig
Carnaval MG	cranberry	large	I	Epamig
BRS Valente	black	small	II	Embrapa
BRS Horizonte	carioca	small	II	Embrapa

<sup>(1)</sup> carioca: beige with light brown stripes; jalo: beige. <sup>(2)</sup> I :determinate growth habit, life cycle between 65 and 75 d; II indeterminate upright growth habit; III: indeterminate semiprostrate to prostrate growth habit. <sup>(3)</sup> Institutions of origin: UFV - Federal University of Viçosa, Embrapa - Brazilian Agricultural Research Corporation, UFLA - Federal University of Lavras, Epamig - Agricultural Research Institute of Minas Gerais.

seedling emergence, plots were sprinkler irrigated with 2.0 cm of water. Then irrigation was applied weekly until the beginning of flowering with approximately 4.0 cm of water. In the reproductive stage, approximately 5.0 cm of water were applied in weekly irrigations.

The canopy closure, seed yield, 100-seed mass, seed Mo concentration (SMoCc), and seed Mo content (SMoCt) were evaluated. Canopy closure was estimated visually in Coimbra at the stages V4, R5, and R7 by observing each plot from one end (looking down the rows) and visually estimating the proportion of soil surface visible between the rows (100 % representing complete soil cover) (Kane & Grabau, 1992). Seed yield and 100-seed mass were estimated

with an approximate moisture content of 13 % (wet basis). Two hundred seeds were oven-dried at 72 °C for 72 h to estimate the 100-seed dry mass. The dried seeds were ground and subjected to nitric perchloric acid digestion, with subsequent analysis in an atomic emission spectrometer with inductively-coupled plasma (ICP-OES, Perkin-Elmer Optima 3300 DV) to determine the SMoCc. The SMoCt represents the amount of Mo accumulated in seed that will be available to the plant grown from it. For its calculation, SMoCc, in µg g<sup>-1</sup>, was multiplied by the average mass of one dry seed, in g.

The data were subjected to analysis of variance, and the means grouped by the Scott-Knott test at 5 %.



## RESULTS AND DISCUSSION

### Canopy closure and yield

In 2010, in Coimbra, the canopy closure of the eight Type III genotypes was 50-60 % in the V4 growth stage, 70-85 % in the R5 stage, and 80-87 % in R7 stage. The canopy closure of Type II cultivars (Pioneiro, Horizonte, and Valente) was 40 or 45 % in the V4 stage, 50 or 55 % in the R5 stage, and 60 or 75 % in the R7 stage. Cultivar Carnaval (Type I) had a canopy closure of 30 % in V4, 45 % in R5 and 50 % in R7. These results clearly indicate that since the V4 stage, when Mo was first sprayed, Type III plants covered more space between rows than either Type II or Type I plants. The characteristics of these three types of plants explain these differences. Type III have more nodes and more lateral branches than Type II plants, while the latter have more lateral branches and more nodes than Type I plants (Vieira et al., 2006).

The average yields in the four experiments were high, ranging from 341 (Oratórios, March 2010, CV = 22 %) to 486 g m<sup>-2</sup> (Oratórios, July 2008, CV = 19 %). In Coimbra, the CV was 14 %; in Oratórios (April 2008) 20 %. Only in Coimbra the effect of genotype on yield was significant, and genotypes were clustered in three groups. The group of highest-yielding genotypes in Coimbra comprised Ouro Vermelho, Pérola, Majestoso, VC-8, Ouro Negro, Madrepérola, Talismã, and Pioneiro.

### Molybdenum concentration and content in the seeds of genotypes without Mo fertilization

The yield in the two 2010 experiments without external Mo application was around 350 g m<sup>-2</sup>. In Coimbra, two yield-related groups were clustered (CV = 14 %, data not shown). In Oratórios (CV = 22 %), genotypes had no significant influence on yield.

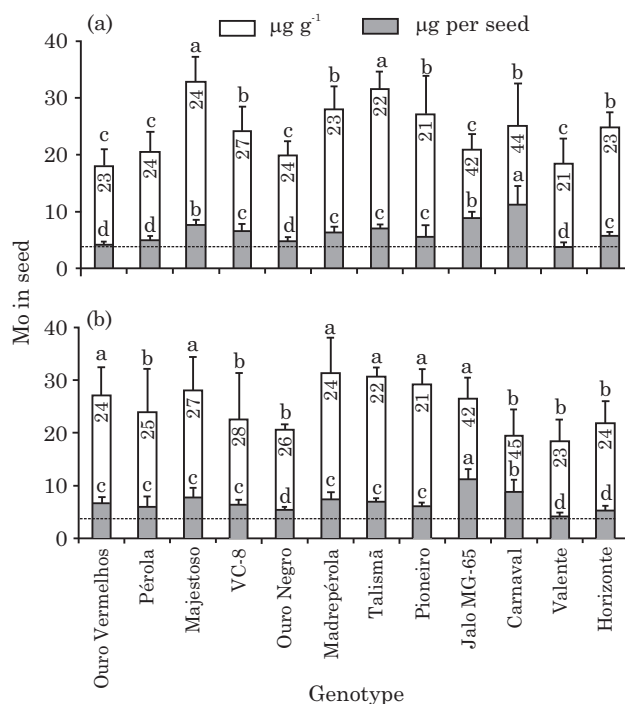
In Coimbra, no Mo was detected in seeds of the genotypes. In Oratórios, Mo was detected in the seeds, but genotypes did not affect SMOcc [F (11, 33) = 0.885, p = 0.56, CV = 14 %]. The average SMOcc in Oratórios was 2.83 µg g<sup>-1</sup>, whereas SMOct varied from 0.61 to 1.32 µg, depending on the genotype (data not shown). These results suggest that the soil in Coimbra is poorer in Mo than the soil in Oratórios, since SMOcc indicates the Mo availability in the soil (Brodrick et al., 1995). According to these authors, the difference among genotypes in the capacity to accumulate Mo in seed is less pronounced in Mo-poor soils, which may be the reason for the absence of a significant effect of genotypes on SMOcc, when no external Mo was used.

### Mo concentration and content in the seeds of Mo-fertilized genotypes

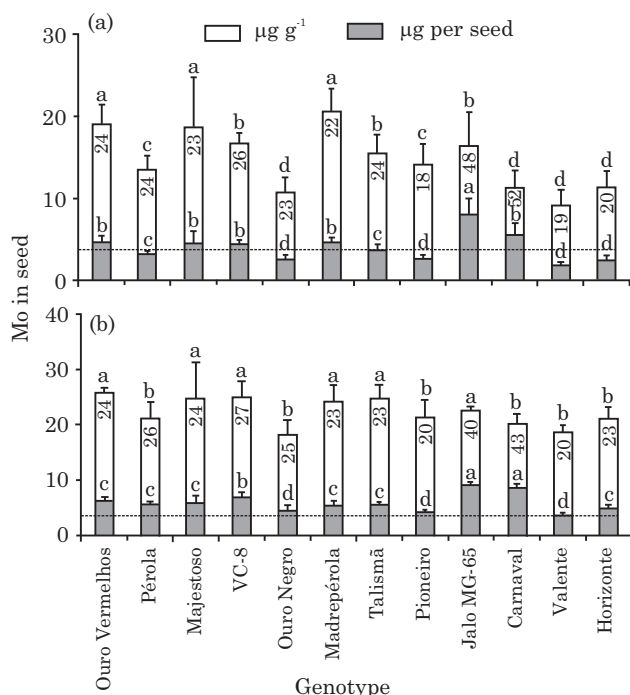
In 2008, the average SMOcc of the genotypes was

24.2 µg g<sup>-1</sup> in the experiment installed in April, and 25.0 µg g<sup>-1</sup> in that installed in July; in 2010, in the experiments installed in March, the average SMOcc was 14.7 µg g<sup>-1</sup> in Coimbra and 22.2 µg g<sup>-1</sup> in Oratórios. The average SMOct was, respectively, 6.39 and 6.84 µg in 2008 and 3.94 and 5.87 µg per seed in 2010. The lowest mean values of SMOcc and SMOct in 2010, especially in Coimbra, reflect the lower soil pH in that year (Table 1). Plants grown in soil at pH 4.7 (Coimbra, 2010) may have less available Mo for uptake than when grown in soil with pH 6.4 (2008), because for every unit increase in pH above 3.0, the solubility of MoO<sub>4</sub><sup>-2</sup> increases 100 times (Lindsay, 1979).

The effect of genotypes on SMOcc and SMOct was significant in all four experiments (Figures 1 and 2). The SMOcc ranged from 9.1 (Valente, Figure 2a) to 32.7 µg g<sup>-1</sup> (Majestoso, Figure 1a). These high values of SMOcc compared to those obtained when no Mo



**Figure 1.** Effect of molybdenum (Mo) sprayed on 12 common bean genotypes at V4 (300 g ha<sup>-1</sup>) and R6 growth stages (300 g ha<sup>-1</sup>) on the Mo seed concentration (□) and content (■) in Oratórios in the experiments of (a) April 2008 and (b) July 2008. The filling of the white bars represents the 100-seed dry mass (in g). The seed Mo content (µg per seed) was calculated by multiplying the mass of one dry seed (in g) by the seed Mo concentration (in µg g<sup>-1</sup>). The horizontal dotted line represents the seed Mo content (3.6 µg) at which common bean does not respond to Mo fertilization (Vieira et al., 2011b). Thin bars represent the standard deviation.



**Figure 2.** Effect of molybdenum (Mo) sprayed on 12 common bean genotypes at V4 (100 g ha<sup>-1</sup>), R5 (250 g ha<sup>-1</sup>), and R7 growth stages (250 g ha<sup>-1</sup>) on the Mo seed concentration (□) and content (■) in the 2010 experiments in (a) Coimbra and (b) Oratórios. The filling of the white bars represents the 100-seed dry mass (in g). The seed Mo content (µg per seed) was calculated by multiplying the mass of one dry seed (in g) by the seed Mo concentration (in µg g<sup>-1</sup>). The horizontal dotted line represents the seed Mo content (3.6 µg) at which common bean does not respond to Mo fertilization (Vieira et al., 2011b). Thin bars represent the standard deviation.

was applied, clearly show that foliar-applied Mo increases the SMOcC substantially, as observed by Vieira et al. (2005, 2010), Pacheco et al. (2012), and Almeida et al. (2013). The SMOcT ranged from 1.75 (Valente, Figure 2a) to 11.2 µg per seed (Jalo MG-65, Figure 1b).

With the exception of Pioneiro (Type II) in 2008 (Figure 1b), the groups with the highest SMOcC were formed of Type III genotypes (Figures 1 and 2). Cultivar Majestoso (Type III) was in the group with the highest SMOcC in all four experiments, and cultivars Ouro Vermelho, Madrepérola and Talismã (Types III) were in this group in three experiments. The greater canopy closure of Type III plants in the growth stages in which Mo was sprayed on foliage may have increased the interception of Mo solution by plants. Pérola, Ouro Negro (Types III), Valente, Horizonte (Types II), and Carnaval (Type I) were never present in the group of genotypes with highest SMOcC.

The absence of some Type III genotypes from the group of genotypes with highest SMOcC indicates that other mechanisms, aside from canopy closure, influenced the SMOcC.

Cultivar Majestoso belonged to a group with higher SMOcC than Ouro Negro and Pérola (Figures 1 and 2), in spite of having been derived from a cross between Ouro Negro and Pérola (Paula Júnior et al., 2010). On the average of the four experiments, Majestoso had a 50 % higher SMOcC than Ouro Negro and 32 % higher than Pérola. The greatest SMOcC difference between genotypes, however, was 61 % (Majestoso vs. Valente). Franco & Munns (1981) and Brodrick et al. (1995) reported a 3.5 and 5-fold difference in SMOcC between genotypes, respectively, although in these studies the Mo sources for the plants were seed and soil. Ouro Negro nodulates well (Paula Júnior et al., 2010), and the nodules are strong Mo sinks (Jacob-Neto et al., 1988; Brodrick & Giller 1991a). Physiologically, the ability of a genotype to accumulate Mo in seeds is associated with its capacity to re-transfer Mo from the roots, nodules and pod walls to the seeds (Brodrick & Giller, 1991b).

Cultivar Jalo MG-65 (Figures 1b and 2a) or Carnaval (Figure 1a), or both (Figure 2b), were in the group of genotypes with the highest SMOcC. These results were expected because the seed mass of these two cultivars is almost twice as high as that of the other genotypes. On average, the SMOcC of Jalo MG-65 (Type III) was 15 % higher than that of Carnaval (Type I). However, because of the higher seed mass of Carnaval relative to Jalo MG-65, the SMOcT of Carnaval was 9 % higher than that of Jalo MG-65. Carnaval is an early-maturing cultivar. In 2008, plants of Carnaval were sprayed in R5 and R7, in later stages than those of the other genotypes (V4 and R6). In that year, Carnaval was not among the genotypes with highest SMOcC. The difference of 7-10 d in the life cycle of Carnaval compared to the other genotypes may have compromised the comparison of SMOcC among genotypes. However, in 2010, when Carnaval was sown a week later than the other genotypes, in order to synchronize the growth stages of all plants, once again Carnaval was not among the genotypes with the highest SMOcC. Therefore, in 2008, if spraying Mo on plants of Carnaval in later growth stages than on plants of the other genotypes caused any bias, the magnitude of this bias was small. The SMOcT of Carnaval and Jalo MG-65 was clearly above 3.6 µg (Figures 1 and 2). Plants grown from seeds with such as high SMOcT do not respond to Mo fertilization in Mo and/or N-poor soil (Vieira et al., 2011b; Almeida et al., 2013).

In one of the experiments in 2010, line VC-8 had the highest SMOcT among the genotypes of Mesoamerican origin (Figure 2b). In the experiment set up in April 2008, Majestoso had the highest SMOcT of these small-seeded genotypes (Figure 1a). In the other two experiments, the SMOcT among cultivars

Ouro Vermelho and Madrepérola was similar to those of line VC-8 and of cultivar Majestoso (Figures 1b and 2a). Majestoso accumulated more Mo per gram of seed tissue than VC-8 in three experiments (Figures 1a,b, and 2a). However, because of the greater 100-seed mass of VC-8 (31.4 g averaged across the four experiments) than that of Majestoso (28.2 g), these genotypes had similar SMOct.

The black bean cultivars Ouro Negro and Valente were always in the group of genotypes with the lowest SMOct (Figures 1 and 2). The Type II cultivars Pioneiro (Figure 2a,b) and Horizonte (Figures 1b and 2a), both of the carioca class, were in this group in two experiments. Cultivars Ouro Vermelho and Pérola were in the group with the lowest SMOct in one experiment (Figure 1a).

On average, the SMOct of Majestoso was 6.4  $\mu\text{g}$  and that of line VC-8, 6.1  $\mu\text{g}$ ; in the other extreme, the SMOct of Ouro Negro was 4.3  $\mu\text{g}$  and that of Valente, 3.4  $\mu\text{g}$ . Therefore, the SMOct of Majestoso was approximately 90 % higher than that of Valente. These results indicate that the Mo rate required to enrich the seeds of Majestoso and VC-8 by plant spraying may be lower than that required to enrich the seeds of Ouro Negro, and especially those of Valente. In common bean genotypes, the effect of the seed coat color on the Mo-accumulation capacity of the seed has yet to be studied in detail. Our results with foliar-applied Mo and the study of Franco & Munns (1981) with Mo supplied via soil may indicate that black seed cultivars, such as, Ouro Negro and Valente, could accumulate less Mo in seeds than genotypes with other seed coat colors.

In Oratórios, all genotypes had SMOct above 3.6  $\mu\text{g}$  (Figures 1a,b, and 2b). In Coimbra, five cultivars did not reach this seed content: Pérola, Ouro Negro, Pioneiro, Horizonte, and Valente (Figure 2a). Plants originated from seeds of these five genotypes grown in Mo-poor soil may respond favorably to Mo application (Vieira et al., 2011b).

Our results suggest that large-seeded cultivars can reach 3.6  $\mu\text{g}$  Mo per seed at Mo rates well below 600  $\text{g ha}^{-1}$ . This Mo rate seems sufficient to increase Mo in seeds of Type III genotypes such as Majestoso, VC-8, Ouro Vermelho, and Madrepérola to 3.6  $\mu\text{g}$ , even in soil with pH 4.7 (as in Coimbra), where the Mo available in soil is 100 times lower than in soil with pH 5.7 (Lindsay, 1979). In a soil with pH 5.2 in Coimbra, Vieira et al. (2010) sprayed plants of cultivar Pérola with 1000  $\text{g ha}^{-1}$  Mo to obtain 3.6  $\mu\text{g}$  Mo per seed. In the present study, Pérola was not among the genotypes with high capacity to accumulate Mo in seed, helping to explain the high Mo rate used by Vieira et al. (2010) to obtain 3.6  $\mu\text{g}$  Mo per seed.

There were some indications that the lower SMOcc in Type I and Type II cultivars was due, at least in part, to the lower interception of the sprayed solution by the crop relative to that of Type III cultivars. However, since Type I cultivars generally have large

seeds, they have higher SMOct values than small seeds. Cultivars with Type II growth habit generally have small seeds. Therefore, our results suggest that, to raise SMOct to 3.6  $\mu\text{g}$ , Type II cultivars require a higher Mo fertilizer rate than both Type I cultivars and III cultivars that are good Mo accumulators in seed.

Canopy closure was only evaluated in Coimbra. In Oratórios, the differences in canopy closure among the cultivars with the three growth habits after Mo application were apparently similar to those in Coimbra. In 2008, plots consisted of one 2-m row. This row was lined on either side by randomly 2 of the 11 other genotypes. Eight of the 12 genotypes were Type III. Plants of these genotypes are more competitive (have more intense growth) than Type I and type II plants. Thus, the probability of Type I and II plants to compete with Types III plants was higher, on either side of the row, than with Type I or Type II plants. Possibly, a bias was introduced by using borders of plants morphologically different from those in the plots. We believe, however, that this bias only had a minor influence on the results of SMOcc in 2008, because at the time Mo application, plants were either young (V4 stage, 50 % of the Mo rate) or had not reached maximum growth (R6 stage). Moreover, the results of SMOcc of the genotypes in 2008 (bordered by other genotypes) were similar to those obtained in 2010 (bordered by the same genotype).

The common bean class carioca (beige with brown stripes) is the most commonly grown in Brazil, followed by the black class. Other relevant classes in certain regions are “manteigão” (all large-seeded genotypes), especially jalo (with beige seeds, e.g., Jalo MG-65), cranberry (e.g., Carnaval), and the red class (Paula Júnior et al., 2010). In Brazil, Type III cultivars are cultivated much more on common bean fields than Type II cultivars. In the case of Type I, few cultivars are available for growers (Vieira et al., 2006). In this study, we used genotypes from four research institutions to represent this variability of commercial classes and plant types.

## CONCLUSIONS

1. Common bean genotypes vary in their capacity of accumulating foliar-applied molybdenum in the seeds.
2. Cultivar Majestoso has a high capacity of accumulating foliar-applied Mo in the seeds, unlike the cultivars Ouro Negro and Valente.

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