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Molybdenum mixed with glyphosate and alone via foliar spray in no-tillage common bean grown on corn stover¹

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ABSTRACT

The effect of molybdenum (Mo) on common bean grown in desiccated corn stover in a no-tillage system was evaluated under two application modes: Mo mixed with the desiccant glyphosate and Mo direct spray to the bean leaves. The treatments (four replicates) were assigned to a completely randomized block design in a split-plot arrangement with the application of Mo (0, 100, 200, 400 and 800 g ha⁻¹) mixed with glyphosate in the main plots and Mo foliar spray (0 and 100 g ha⁻¹) in the sub-plots. The field experiments were carried out in 2009 and 2010 in the municipality of Coimbra, Minas Gerais State, with the common bean cultivar Ouro Vermelho. Mo mixed with glyphosate had neither an effect on common bean yield nor on the Mo and N contents in leaves, however it increased the Mo and N contents in seeds. Application of Mo via foliar spray increased Mo content in leaves and Mo and N contents in seeds. The reapplication of molybdenum with glyphosate for desiccation in subsequent crops caused a cumulative effect of Mo content in bean seeds.

Key words: *Phaseolus vulgaris* L., desiccation, molybdenum fertilization.

RESUMO

Molibdênio aplicado com glifosato e isolado por via foliar em feijoeiro cultivado sobre palhada de milho no sistema de plantio direto

O objetivo deste trabalho foi estudar o efeito da aplicação do molibdênio (Mo), em mistura com dessecante, em feijoeiro cultivado sobre palhada de milho (*Zea mays*), no sistema de plantio direto. Utilizou-se o delineamento em blocos casualizados, com quatro repetições, no esquema de parcelas subdivididas. Nas parcelas, foram aplicadas doses de Mo (0, 100, 200, 400 e 800 g ha⁻¹), juntamente com herbicida dessecante (Glifosato) e, nas subparcelas, duas doses de Mo (0 e 100 g ha⁻¹), por via foliar. O trabalho foi realizado no município de Coimbra, MG, nos anos de 2009 e 2010, utilizando-se o cultivar Ouro Vermelho. A aplicação das doses do molibdênio, em operação de dessecação, com glifosato, em sistema de plantio direto, não influenciou a produtividade de grãos e o teor foliar de molibdênio e de nitrogênio nas plantas de feijão; porém, elevou os teores de molibdênio e de nitrogênio nos grãos do feijoeiro. Os

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teores de molibdênio nas folhas, bem como os teores de molibdênio e de nitrogênio nos grãos do feijoeiro aumentaram com a aplicação do molibdênio, isolado, por via foliar. A reaplicação do molibdênio, em operação de dessecação com glifosato, na mesma área, em cultivos subsequentes, proporcionou efeito acumulativo no teor de molibdênio nos grãos de feijão.

Palavras-chave: *Phaseolus vulgaris* L., herbicida, adubação mólíbdica.

INTRODUCTION

The increase in common bean (*Phaseolus vulgaris* L.) yield and therefore the decrease of the relative cost of micronutrient application, such as Mo, mainly because of its participation in symbiotic nitrogen fixation and nitrate reductase structure, as well as the expected gains in scale, have prompted research and application of this element in common bean fertilization (Berger *et al.*, 1996; Pessoa *et al.*, 2001; Ascoli *et al.*, 2008).

Although common bean is capable of establishing symbiotic associations with rhizobia and fixing atmospheric nitrogen (N_2), the crop depends on supplemental nitrogen fertilizer to achieve substantial yields. In Brazil, the deficiency of nutrients in the soil, especially nitrogen (N), is a major determinant of low bean yields. Because of the high cost of nitrogen fertilizers and the low purchasing power of most farmers, it is necessary to provide less costly alternatives, especially for low-income farmers (Leite *et al.*, 2007).

Nitrogen is the essential nutrient absorbed in the largest quantity by the bean crop and is required in amounts above 100 kg ha⁻¹ to ensure high yields (Fernandes *et al.*, 2005). The application of foliar Mo is reported as effective in reducing the amount of nitrogen fertilizer to be used in the bean crop, since Mo is essential for the nitrogen metabolism in the plant. Mo participates as a structural element of the nitrogenase enzyme responsible for the symbiotic fixation of atmospheric nitrogen (N_2) to ammonia (NH_3) by *Rhizobium*, and also the nitrate reductase, the first step in the N incorporation into organic molecules (Marschner, 1995).

According to Wichard *et al.* (2009), the binding of Mo to organic matter helps stop leaching of the nutrient and is considered critical for fixing N in the ecosystems. In litter, Mo is strongly complexed with tannins derived from plants in a wide pH range and, in deeper soils, Mo binds both to iron oxides and organic matter (Wichard *et al.*, 2009). These authors also suggested that organic-matter bound Mo can be captured by complexing agents released by N_2 -fixing bacteria.

No-tillage (NT) is a very important alternative in grain production that allows cultivation without major negative impacts on the environment and contributes to a greater

conservation of soil and water resources. However, it is dependent on the quantity, quality and permanence of crop residue on soil surface, which is met by corn stover. The amount of crop residue provided by corn after export of grain has been enough to ensure efficient soil cover (Gomes Júnior *et al.*, 2008), besides being a good rotation crop with legumes, such as common bean.

Although there is no response pattern to Mo, the increase in yield of the bean crop with supply of Mo has been reported in a number of studies in field conditions (Araújo *et al.*, 2009; Nascimento *et al.*, 2009; Biscaro *et al.*, 2011; Rocha *et al.*, 2011). Studies prove the feasibility of fertilizer foliar application on V4 stage bean, mainly of micronutrients tank-mixed with herbicides and pesticides to reduce costs and optimize equipment usage (Araújo *et al.*, 2008; Silva *et al.*, 2003b). In the experiment of 2008, Damato Neto (2010) found increased Mo content in bean seeds, but with no change in yield after application of Mo mixed with the desiccant glyphosate on corn stover in no-till.

There is evidence that the common bean is capable of absorbing the Mo applied to corn stover and mixed with desiccant, but there is no information about the crop performance over the years. Thus, the 2008 experiment of Damato Neto (2010) was repeated for two more years, in 2009 and 2010, to study the effect of different Mo rates applied in the desiccation with glyphosate on corn stover and alone via foliar spray in no-till bean.

MATERIAL AND METHODS

The two experiments conducted in 2009 and 2010 were carried out in the experimental station of the Department of Plant Science of the Universidade Federal de Viçosa located in the municipality of Coimbra, Minas Gerais. The geographic coordinates of the area are 20° 50' 30" S and 42° 48' 30" O, 715 m altitude, in a dystrophic Red Yellow Podzolic soil of clay texture, with 540, 180 and 280 g kg⁻¹ of clay, silt and sand respectively (Embrapa, 2006).

For cultivation of no-tillage common bean on corn (*Zea mays*) stover in second and third years, the corn crop was planted in November 2008 and 2009, respectively. The first experiment was established by sowing corn in December 2007 and sowing bean in May 2008 (Damato Neto, 2010).

The subsequent experiments with bean were installed in the same field and the treatments were randomly assigned to plots. Before applying the treatments to the first bean crop (2008), soil samples were collected at a depth of 0-20 cm for the chemical analysis (Table 1). Common bean cultivar Red Gold (indeterminate growth, plant type II/II) was sown (15 seeds m^{-2}) in March 26, 2009 and April 14, 2010 (autumn-winter planting), using a seeder-fertilizer machine with double disc furrow opener with offset discs.

The treatments (four replicates) were assigned to a completely randomized block design in a split-plot arrangement with the application of Mo (0, 100, 200, 400 and 800 g ha^{-1}) mixed with glyphosate (isopropylamine salt - Roundup Original® - 1440 g e.a. ha^{-1}) in the main plots and Mo foliar spray (0 and 100 g ha^{-1}) in the sub-plots.

The plots consisted of four rows of bean plants (10 m long and 0.5 m apart) divided into subplots of 5 m long rows. The harvest area of the subplot was delimited by the two central rows (4 m^2), excluding 0.5 m on each end of the rows.

In both years, 13 days before sowing, desiccation of weeds along the corn stover was done with glyphosate mixed with Mo (sodium molybdate - Na_2MoO_4 - with 39% Mo) using the rates mentioned above. The application was performed using a CO_2 -pressurized backpack sprayer (2 bar), equipped with a 2-m spray bar, TT 11002 flat fan nozzles spaced 50 cm which delivered a spray volume of 140 L ha^{-1} .

The starter fertilizer was placed in furrow, using 350 kg ha^{-1} of the 8-28-16 (N- P_2O_5 - K_2O) formulation. Foliar fertilization (Na_2MoO_4 with 39% Mo) was sprayed on V4-stage bean plants, using a backpack sprayer with pressure relief valve (2 bar) (VPC azul-465344; Jacto) equipped with a 2-m spray bar, TT 11002 flat fan nozzles spaced 50 cm which delivered a spray volume of 180 L ha^{-1} .

In both experiments, the nitrogen fertilization was carried out 28 days after seedling emergence (DAE) by hand application of 45 kg ha^{-1} of N as urea, next to the row on the soil surface. After N application, irrigation was applied (under 10 mm of water layer) to minimize losses by volatilization.

Weed control in the bean crop was carried out with a commercial mixture of herbicides fluazifop-p-butyl + fomesafen (200+250 g L^{-1} , respectively) at the commercial rate of 0.8 L ha^{-1} , in both years. If necessary, plants were sprayed with the fungicide azoxystrobin (120 g ha^{-1}) to

control rust [*Uromyces appendiculatus* (Pers.) Unger] and angular leaf spot [*Phaeoisariopsis griseola* (Sacc.) Ferraris] and Fluazinan for white mold (*Sclerotinia sclerotiorum*). Insect control with deltamethrin (80 ml ha^{-1}) was applied when the pest population was moderate. The crop was kept under sprinkler irrigation. The soil water content was maintained near field capacity.

SPAD index readings (Soil Plant Analyses Development) were measured using a Minolta SPAD-502 chlorophyll meter on third fully expanded trifoliate leaves, from the apex, in ten R5-stage plants randomly selected within the harvest area of the subplot, always between 9:00 and 10:00 am. The average of the readings of the ten sampled plants represented the value of the subplot. These leaves were also used for analyses of N and Mo.

After the physiological maturity stage, plants were harvested from the subplot and left in the sun to finish drying. The following variables were determined: final stand (plants ha^{-1}), pod number per plant, seed number per pod, 100-seed mass and seed yield. Seed yield (kg ha^{-1}) was calculated from the production of the harvest area of each subplot, standardizing the moisture content (wet basis) at 13%. Seed samples were also collected from the harvest area of each subplot.

Leaf and seed samples of bean were ground and mixed for nitroperchloric digestion and Mo was determined by inductively coupled plasma optical emission spectrometry using a Perkin Elmer Optima 3300 DV simultaneous spectrometer. Leaf nitrogen content was calculated by summing the organic-N contents determined (after sulfuric acid digestion) by the Nessler's reagent colorimetric method and the nitrate-N contents (Malavolta *et al.*, 1997). Seed nitrogen content was determined by the micro-Kjeldahl method (Malavolta *et al.*, 1997).

For statistical analysis we used individual and combined analysis of variance, with the SAS Institute 9.1 for Windows (SAS, 2003). Means of the treatments with and without Mo application were compared by the F test. The models for the quantitative factors (rate of Mo mixed with glyphosate) were chosen based on the significance of the coefficients of regression (t test at 5% probability) and determination ($R^2 = SS \text{ Regression} / SS \text{ Treatment}$), as well as the inherent characteristics of the biological phenomenon under study.

Table 1. Soil chemical characteristics* at 0-20 cm depth in the experimental area, Coimbra – MG

pH	P	K ⁺	H + Al	Al ³⁺	Ca ²⁺	Mg ²⁺	CTC _{total}	CTC _{effective}	SB	V	M	P-rem
H ₂ O	mg dm ⁻³				cmol _c dm ⁻³					%		mg L ⁻¹
6.1	7.4	95	3.8	0.0	2.7	1.1	7.8	4.0	4.0	52	0.0	24.5

* P and K - Extractor: Mehlich 1; Ca, Mg and Al - Extractor: KCl 1 mol L^{-1} ; H + Al - Extractor: Calcium Acetate 0.5 mol L^{-1} , pH 7.0.

RESULTS AND DISCUSSION

There was no effect of Mo rates mixed with glyphosate applied for desiccation on the final stand of beans grown on corn stover. However, in the evaluation of the interaction of Mo rate applied by foliar spray and the year, with or without Mo application, the number of plants per hectare in the final stand in 2009 was higher than in 2010 (Table 2). It is noteworthy that in 2009, with the foliar application of Mo, the final stand had higher number of plants per hectare than those not fertilized with Mo (Table 2).

There was no effect of Mo rates mixed with glyphosate on pod number per plant (PNP) (mean = 10.24 un.; Standard deviation = 0.92 un.). This result was similar to that reported by Barbosa *et al.* (2010) with different N rates, in the presence or absence of 80 g ha⁻¹ Mo, finding no significant influence on PNP in two years of winter bean cultivation. Fernandes *et al.* (2005) evaluated the application of foliar Mo and N, in-furrow or sidedress, to no-tillage bean on corn stover and also found no difference for PNP. However, reports of increased PNP in response to Mo application are common in the literature (Pessoa *et al.*, 2001; Silva *et al.*, 2003a; Nascimento *et al.*, 2009). For the year effect, the average PNP was higher in 2010 (Table 3). The probable reason for this fact is the plasticity of bean plants, which are able to compensate the primary yield components in different environmental conditions, including the plant population (Piana *et al.*, 2007), which in 2010 was lower than in 2009 (Table 2).

Rates of Mo mixed with glyphosate applied for desiccation and rates of Mo applied by foliar spray did not influence the seed number per pod (SNP) (mean = 6.12

un.; standard deviation = 0.35 un.) and the 100-seed mass (100SM) (mean = 25.64 g; standard deviation = 0.66 g). However, there was significant difference in the SNP between the years of cultivation; in 2009, the average was lower than in 2010 (Table 3), which was similar to result reported by Ascoli *et al.*, (2008). Nevertheless, Leite *et al.*, (2007) observed increase in these yield components (SNP; 100SM) with the application of foliar Mo to common bean.

Mo rates mixed with glyphosate had no significant effect on SPAD readings; means ranged from 39.5 to 41.2 SPAD un.. These results were different from those found by Pires *et al.* (2004) and Ferreira *et al.* (2002), who found increase in SPAD index of 29 and 37%, respectively, for common bean fertilized with molybdenum. However, we found difference in the SPAD index reading between the two years. In 2009, the average was higher than in 2010 (Table 3). Ferreira *et al.* (2002) also found variation in SPAD index readings from one year to another, using the same cultivar.

Seed yield (SY) was not influenced by the combination of different rates of Mo with glyphosate (mean = 2772.2 kg ha⁻¹; SD = 200.7 kg ha⁻¹). The coefficients of variation (CV %) in the plot and subplot were respectively, 12.7 and 7.2. These CV values are below acceptable limits for the common bean (Oliveira *et al.*, 2009). It is important to note that the experiment had an adequate degree of precision, reflected in the low CV values reported for the other yield components (Table 3).

The lack of yield response may be related to the concentration of the nutrient in the soil-plant system. The soil pH (6.1) in the area where the experiments were installed (Table 1) may have contributed to the increased availability of Mo, providing the adequate amount for the

Table 2. Unfolding of the significant interactions in the analysis of variance for final stand and leaf Mo content (LMoC) of common bean grown on corn stover, in 2009 and 2010, Coimbra – MG

Mo Foliar g ha ⁻¹	Final stand plants ha ⁻¹		LMoC mg kg ⁻¹	
	2009	2010	2009	2010
0	178625 Ab	166000 Ba	0.41 Ab	0.43 Ab
100	193000 Aa	164750 Ba	1.87 Ba	3.67 Aa

Means followed by the same capital in the row and small letter in the column are not significantly different by the F test at 5% probability.

Table 3. Pod number per plant (PNP), seed number per pod (SNP) and SPAD Reading (SPAD) of no-till common bean grown on corn stover, in 2009 and 2010, Coimbra – MG

Year	** PNP		**SNP		*SPAD	
	2009	2010	2009	2010	2009	2010
Units						
Mean	9.85 B	10.63 A	5.90 B	6.35 A	41.0 A	39.9 B
CV (%) of plot	10.5		6.9		4.6	
CV (%) of subplot	9.0		5.7		4.0	

Means followed by the same capital letter in the row are not significantly different by the F test; at * 5% probability and ** F 1% probability levels

plant to complete its life cycle. In soils with relatively high pH (greater than 6.0), response to the application of Mo is not usually expected, because there is increased availability of the micronutrient with increasing pH (Silva *et al.*, 2012). Another reason for the lack of difference between yield means is that the application of 45 kg ha⁻¹ sidedress N in the form of urea and the amount of N released by microbial mineralization of organic matter have supplied bean plants.

Calonego *et al.* (2010) evaluated the effect of five N rates (25, 50, 75, 100 and 125 kg ha⁻¹) applied 20 days after emergence (DAE) associated with and without foliar application of 80 g ha⁻¹ Mo at 25 DAE and found that the molybdenum fertilization increased bean yield regardless of sidedress N rates. In a similar study, Biscaro *et al.* (2011) found that sidedress N increased the seed yield of common bean only when combined with foliar Mo.

Leaf molybdenum content (LMoC) was not influenced by the rates of Mo mixed with glyphosate, with means (two years) ranging from 1.26 to 2.09 mg kg⁻¹. However, these means showed a trend of linear increase with increasing rates of Mo mixed with glyphosate. Failure to statistically detect differences among the LMoC means, as a function of Mo rates mixed with glyphosate, was probably due to the high CV% of the data (Table 4). Other studies reported increased LMoC in beans with Mo supplementation (Ferreira *et al.*, 2003; Pires *et al.*, 2005). When comparing the years, LMoC was higher in 2010 (Table 4).

Foliar application of Mo influenced LMoC. The average LMoC of treatments that received Mo supplementation (100 g ha⁻¹) was 2.77 mg kg⁻¹, while the average LMoC of treatments without additional Mo was 0.42 mg kg⁻¹. In the interaction Mo foliar spray and year, regardless of the year, the lowest means of LMoC occurred in subplots without foliar application of Mo (Table 2). The subplots with foliar application of Mo had LMoC means higher in 2010 (Table 2). It stands out that, in both years, the LMoC means recorded in plots without foliar Mo were within the sufficiency range for LMoC (0.40 to 1.40 mg kg⁻¹) proposed for the common bean by Oliveira & Thung (1988). That is, the lack of

Mo fertilization was not limiting for the development of the legume.

There was no effect of application of Mo rates mixed with glyphosate on leaf N content (LNC) of no-till beans grown on corn stover. Means ranged from 5.04 to 5.34 dag kg⁻¹. The LNC found in this study are greater than those reported by Martinez *et al.* (1999) as the sufficiency range of N for the common bean (3.0 to 3.5 dag kg⁻¹).

Seed Mo content (SMoC), in the interaction of Mo rates mixed with glyphosate x year (Figure 1), increased with the rate of Mo mixed with the desiccant applied on corn stover, regardless of the year. However, seeds harvested in 2010 had higher average SMoC, which shows the ability of bean plants to absorb and accumulate Mo in the seeds, even when applied with glyphosate at the time of desiccation. This ability, according to Jacob Neto & Rossetto (1998), is an advantage for seed production, since seed reserves of molybdenum are usually sufficient for the plant to grow without depending on Mo supplementation. There is evidence that Mo content exceeding 1.272 µg per seed would benefit plants with regard to leaf N content and yield (Vieira *et al.* 2005).

The application of Mo alone, by foliar spray, increased SMoC. In the treatments without foliar application of Mo, the average SMoC was 1.75 mg kg⁻¹, while in the subplot with additional application of 100 g ha⁻¹ Mo, the average SMoC was 4.27 mg kg⁻¹. For the year effect, similar to what happened to LMoC, the SMoC in 2010 was higher than the average of 2009 (Table 4).

There was no effect of application of Mo rates mixed with glyphosate on seed nitrogen content (SNC). Means were not significantly different ($p < 0.05$), but the data did not fit the regression models. The plots without application of Mo mixed with glyphosate had average SNC of 3.62 dag kg⁻¹, or 5% lower than the average (3.80 dag kg⁻¹) of those applied with Mo mixed with glyphosate (Table 5). For the year effect, in 2009 the average SNC was lower than the average of 2010. This result may be due to the use of Mo accumulated in the soil, from applications in the previous years (Table 4). The practical application of this information is that it is possible to reduce the use of Mo mixed with the desiccant over the years.

Table 4. Leaf molybdenum content (LMC), seed molybdenum content (SMoC) and seed nitrogen content (SNC) in no-till common bean grown on corn stover, in 2009 and 2010, Coimbra – MG

Year	LMoC		SMoC		SNC	
	2009	2010	2009	2010	2009	2010
	kg ⁻¹				dag kg ⁻¹	
Mean	1.14 B	2.05 A	2.72 B	3.30 A	3.51 B	4.03 A
CV (%) of plot	88.7		22.5		4.8	
CV (%) of subplot	86.2		18.1		3.5	

Means followed by the same capital letter in the row are not significantly different by the F test; at 1% probability level.

Seed N content (SNC), in the interaction of Mo rates mixed with glyphosate x Mo foliar spray (Figure 2), remained constant when 100 g ha⁻¹ of Mo were applied on bean leaves. However, without foliar Mo, there was an exponential response of SNC means that required a rate of 199 g ha⁻¹ Mo mixed with glyphosate to equal the average N content obtained with the foliar application of Mo (Figure 2). These results showed that bean plants absorbed and translocated the Mo

applied with glyphosate to the seeds, making it unnecessary its application via foliar spray under these conditions.

There was significant effect of Mo application to common bean plants via foliar spray on SNC. The subplots without application of Mo via foliar spray had average SNC equal to 3.72 dag kg⁻¹, which was lower than the average of the subplots with the additional application of 100 g ha⁻¹ Mo, 3.81 dag kg⁻¹.

Table 5. Means for seed N content (SNC) of no-till common bean grown on corn stover as a function of Mo rates mixed with glyphosate in 2009 and 2010, Coimbra MG

Mo g ha ⁻¹	1,440 g e.a. ha ⁻¹ Glyphosate + Mo rate					Mean
	0	100	200	400	800	
SNC (dag kg ⁻¹)	3.62	3.82	3.73	3.84	3.82	3.77

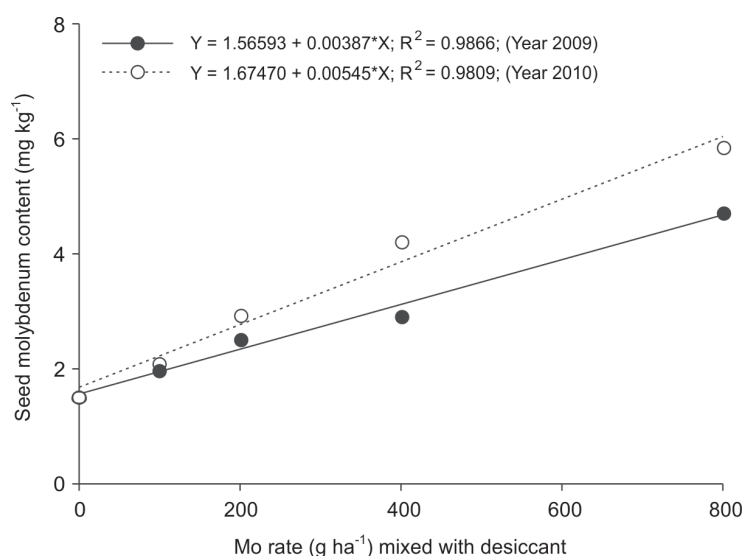


Figure 1. Seed Mo content of common bean as a function of Mo rates mixed with glyphosate in 2009 and 2010, Coimbra - MG.

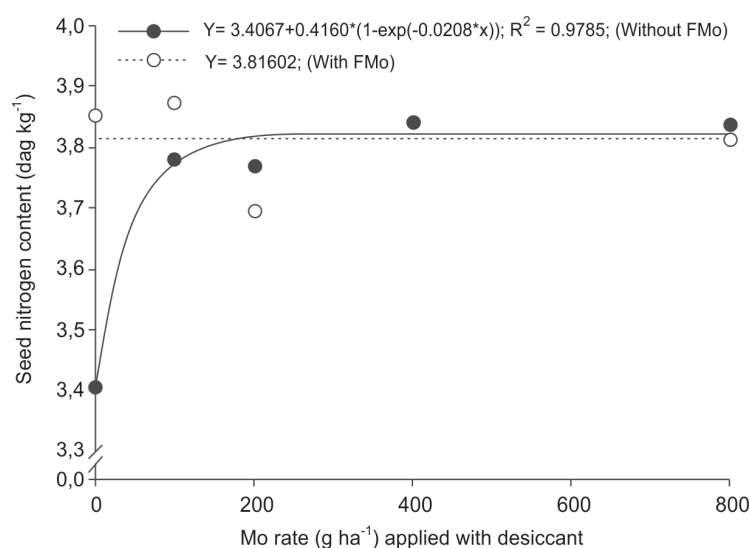


Figure 2. Seed N content of common bean, with or without foliar Mo application as a function of Mo rates mixed with glyphosate, Coimbra - MG.

CONCLUSIONS

The application of molybdenum rates mixed with glyphosate for desiccation in no-tillage system did not affect seed yield and leaf content of molybdenum and nitrogen in bean plants, but increased the contents of molybdenum and nitrogen in the seeds. The molybdenum content in leaves as well as the molybdenum and nitrogen contents in bean seeds increased with the application of molybdenum via foliar spray. The re-application of molybdenum mixed with glyphosate for desiccation in subsequent crops caused a cumulative effect of molybdenum content in common bean seeds.

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