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EFFICIENCY OF FOLIAR SULFUR SUPPLY TO SOYBEAN IN DIFFERENT DEVELOPMENTAL STAGES

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ABSTRACT

The soybean is a crop with world importance, which grains have several usages in the human and animal nutrition. Among the essential nutrients the sulfur is a macronutrient constituent of proteins and amino acids and is required in appreciable amount by the leguminous crop. The objective of this work was to investigate the effect of sulfur, by foliar application in different developmental stages of soybean, under vegetative characteristics and yield components. The experiment was carried out under field conditions by completely randomized blocks design with four replications, in factorial scheme 3x5 (3 times of foliar application and 5 doses of sulfur). The TMG132RR soybean cultivar was used in this investigation with the doses of 0, 250, 500, 750 or 1000 mL ha^{-1} of sulfur applied in the stages V2, V2+10 or V2+20 days. The time of foliar application of sulfur has influenced the dry matter of aerial part and the plant height, being the stage V2+20 days the most recommended time. The response to the different doses of sulfur varied based on the results of foliar chlorophyll rate and plant height. For the grain yield there was interaction between time of application of sulfur and dose, being the dose of 731 mL ha^{-1} applied in V2 stage as the maximum yield of 3,690 Kg ha^{-1}. The yield components number of pod per plant and number of grain per pod were not affected by the times or doses of sulfur application. For thousand grain weight the responses to the different doses varied as the time of application.

Keywords: *Glycine max.* Mineral nutrition. Crop Management. Yield Components

1 Introduction

The soybean (*Glycine max*) is a crop of world agricultural importance. In 2019 the USA had the greater mean yield of the grain in the world, with about 3,468 kg ha^{-1}, followed by Brazil with mean of 3,201 kg ha^{-1}. The Mato Grosso state is the major soybean producer in the country. In the 18/19 crop the state production reached 32,454.5 millions of tons, in a total area of 9,699.5 millions of hectares, but despite the large soybean production in Mato Grosso the yield is at levels below the national mean, with 3,120 kg ha^{-1} [1].

One of the factors influencing the soybean yield is the deficiency of some essential nutrients, like the sulfur, which is the fourth main nutrient required by the crop. The need of sulfur in mineral nutrition of plants is already known for much time, mainly due to its participation in enzymes and protein formation. In general, soils cultivated for many years with continuous usage of concentrated fertilizers, hence with low levels of sulfur, end up generating deficiency of this element on the agricultural crops, mainly in the Cerrados region of Brazil [2].

The sulfur deficiency in Brazilian soils is influenced by several factors, the low fertility, added to the low levels of organic matter, the increase in nutrient exportation due to high yields and the high sulfate leaching are the main causes. Richart et al. [3] did not observe increase in sulfur levels in the layer of 0 to 10 cm depth after applications until 60 kg ha^{-1} of sulfur and attribute this fact to the facility of removing sulfate ion on the soil profile. The authors still highlight more attention should be given to sulfur in order to it does not become limiting to the crops, mainly in soils with low levels of organic matter. In this way, the adequate sulfur level in the soil is a factor determining the growth and yield of crops. Soils deficient in sulfur generate grains of lower quality, especially in terms of protein formation, besides take the plant more vulnerable to diseases and abiotic stress [4-6].

Currently, the soil supply of nutrients is one of the main practices that ensure higher yield levels of the crops. However, the application of nutrients via soil is one of the more costly practices in the cost of agricultural products, due to price volatility and its impacts on the profit of the crops. The sulfur absorption occur on the sulfate form S-SO_4^{2-}, it can also be absorbed as organic (S), SO_2 (air) and wettable S (pesticides) via foliar. On the plant it presents itself on the organic form on the majority (cystine, cysteine, methionine, protein, glycosides and vitamins). It is found on the literature works associating the sulfur assimilation with the nitrogen assimilation, being one correlated with the other and with the levels of chlorophyll in leaves and grain yield [7, 8].

Sulfur supply can be raised by increasing organic matter, or supplying fertilizers rich in this nutrient. The inputs with the presence of sulfur are: plaster, simple superphosphate, elemental sulfur (S^0) and formulated with the sulfur addiction. In Brazil, little is known about the usage of sulfur and the research results are not frequent in the country. It happens, in part, due to the nutrient isolation that is complex, requiring the use of pure products, such as elemental sulfur, and in most cases the applications occur through compound products. Besides that, the most technician, agronomists and farmers still consider the sulfur as a secondary element [9]. The sulfur is, probably, the macronutrient less employed on fertilizations and little studied [10].

The Embrapa Soja [11] determined the necessity of nutrients for the production of one ton of soybean grain: 83 kg of N; 15.4 kg of P; 38 kg of K; 12.2 kg of Ca; 6.7 kg of Mg; 15.4 kg of S; 77 g of B; 515 g of Cl; 26 g of Cu; 460 g of Fe; 130 g of Mn; 7 g of Mo e 61 g of Zn. However, this data are for the whole Brazil, there is a lot of misinformation about the real sulfur necessity in the Mato Grosso crop system, as well as the yield losses that are happening. So, it is increasingly necessary to perform researches that assess the efficiency of sulfur fertilizers, namely, know the real sulfur gains at the crops, as well as the best doses for each soybean cultivar, the distribution (times/uniformity of application) of the element in the soil-plant system to understand the real necessity and importance of sulfur on the agricultural productivity. That said, the objective of the work was to determine the effect of sulfur usage, about vegetative characteristics and yield components, by foliar application in different stages of soybean development.

2 Material and Methods

The experiment was carried out in commercial area, cultivated in minimum tillage system for five years, between 2018 October and 2019 January, on Sinop-MT city. The site was georeferenced (latitude: 11º51'57'05'' S; Longitude: 55º23'51'' W) with 380m altitude in flat topography. The climate, according to Koppen-Geiger, is classified as Aw, having two well definite seasons, being one rainy between October to April, and the other dry between May to September, with low annual thermal amplitude, varying from 24 to 27ºC and with annual mean rainfall about 2,100 mm [12].

The accumulated precipitation during the experiment, between 2018 October to 2019 February, was about 1,179.85 mm, a volume larger than the soybean requirements, which is about 450 to 800 mm per cycle, and enough for soybean reach high yields. The registered temperatures in the period were not limiting for growth, development and yield.

It was made, with the help of a probe, the soil sampling in the experimental area, in the layer of 0 to 20 cm depth. Five samples was collected (sub samples), which after joining gave rise a compound sample. The sample was sent to accredited laboratory for chemical analyze with the following results: pH ($CaCL_2$) 5.4; M.O. 18.55 g dm^3; macronutrients: P (Melich) 6.07 mg dm^3; K 52.00 mg dm^3; Ca 2.84 mg dm^3; Mg 0.93 mg dm^3; S 4.0 mg dm^3; V= 57,2%; Ca/Mg relation 3.05; Ca/K; 21.85; Mg/K; 7.16. The micronutrients in mg dm³ were: Zn 5.51; Cu 0.44; Fe 199.16; Mn 11.25; B 0.15. The physical analyze revealed the levels of 497 g dm^{-3} of sand; 125 g dm^{-3} of silt and 378 g dm^{-3} of clay. The local soil is classified as red yellow latosol [13].

Based on the soil analyze result we can observe that the base saturation value is near from the requirement of the crop (60%), not demanding liming. The fertilization was made by hauling in pre-planting providing NPK 00:18:18 500 kg ha^{-1}, according to the farmer management, providing phosphorus and potassium to the soybean, as recommendations of Sousa and Lobato [14].

The experimental design was completely randomized blocks with four replications, in a factorial scheme 3x5. The treatments consisted of 3 times of application: vegetative V2 stage, V2 plus 10 days and V2 plus 20 days, which coincided with the flowering time; and 5 doses of sulfur: 0, 250, 500, 750 and 1,000 mL ha^{-1}, totaling 60 experimental plots. The sulfur was purchased commercially (S-MAX®), having sulfur level of 50%.

The experimental plots were made up for five rows of five meter long, totaling 12.5 m^2. The useful area of the plot was considered the three central rows with four meters long, totaling 6 m^2. The end rows were considered as border, discarding yet a half meter in the edge of each central row. The early soybean cultivar TMG 132RR was sowed at the density of 15 plants m^{-1}, aiming to obtain, after thinning, a density of 260,000 plants ha^{-1}. The soybean cultivar has determined growth type, hilo color light brown, tolerant to lodging, cycle of 118 to 122 days, high demanding in soil fertility to express higher yields and is indicated for planting between October 10 to November 10 at the region.

Before the sowing the seeds were treated, applying Fipronil based insecticide, from the pirazole group (Regent®), and the fungicides Piraclostrobina, from the group strobilurin, and Methyl thiophanate (Standak Top®), from the group of benzimidazol, at the dose of 2 mL kg^{-1} of seeds. The micronutrients cobalt and molybdenum were also applied at the dose of 5 g of Co ad 42 g of Mo, to increase the nodulation efficiency. Before the sowing, the seeds were inoculated with peat soybean inoculant, *Bradyrhizobium japonica,* strain SEMIA 5079 and 5080, minimum concentration of rhizobium of 7 x 10^9 cells/g and dose of 200 g ha^{-1}. Also was employed, in the liquid form, dose of 200 mL ha^{-1} with rhizobium concentration of 5 x 10^9 cells/mL, *Bradyrhizobium elkanii*, strain SEMIA 587 and 5019. The cultural management was made accordingly the crop requirements (Embrapa, 2011). For rust foliar control it was made four applications of fungicides from the strobilurin and triazole group. The weed control in post emergence occurred inside the recommended period, from germination up to 30 days after sowing. Before sowing, weed desiccation was carried out applying 1.5 kg ha^{-1} of glyphosate (granulated) and in post emergence, at 30 days after emergence, it was applied again 1.5 kg ha^{-1} of glyphosate, using a spray volume of 100 L ha^{-1}.

At full flowering soybean stage we evaluated the foliar chlorophyll level, sampling in each plot six intact leaves from six different plants in the middle region. For this, we used a chlorophyll meter of the ClorofiLOG® brand (model CFL-1030), which estimates the chlorophyll level by indirect form, by means of clorofiloG units. Still in full flowering stage we evaluated the number of leaves per plant, shoot dry weight and plant height, sampling four plants per plot. The shoot dry weight was obtained keeping the samples in paper bags and leaving them to air forced oven at 65ºC until constant weight. The number of leaves was counted in four plants per plot. For plant height we measured the plants from soil until the last leaf in apex and obtained the mean value of four plants.

The harvest was handmade on 2019 February 4th. After the harvest, the number of pods per plant and number of grain per pod were counted in four plants per plot, soon later the grains were threshed mechanically. After threshing the grains were clean and sieved by hand and conditioned in paper bags identified. The grain moisture was corrected for 130 g kg^{-1} of water in air forced oven at 60ºC. Later, the thousand grain weight (g) and the grain yield (kg ha^{-1}) were determined. The initial level of water in the grains was determined by direct way, in air forced oven at 105ºC for 24 hours.

The data was submitted to analysis of variance (ANOVA) at the level of 5% probability, by the F test, with the help of SISVAR statistical software [15]. The means were compared by the Scott-Knott test at 5% probability.

3 Results and Discussion

The characteristics evaluated at the flowering stage, namely, chlorophyll level and number of leaves per plant, were not affected by the different times of sulfur application. For shoot dry weight and plant height there were significant differences for times of sulfur application, being the V2+20 days stage (flowering) the one that caused the highest values, values similar to the ones obtained by Pereira et al. [16] (Table 1). For this time of sulfur application the shoot dry weight mean was about 9% superior than the mean of the other times of application. For plant height this superiority was about 12%.

Table 1- Means of the variables evaluated at the full flowering: Chlorophyll level (CLO), number of leaves per plant (NLP), shoot dry weight (SDW) and plant height (PH) on soybean in function of times of sulfur application

Times of application	CLO (un. ClorofiloG®)	NLP	SDW (kg)	PH (m)
V2	43.20 a*	18.50 a	22.71 b	0.74 b
V2+10 days	43.00 a	18.48 a	22.92 b	0.76 b
V2+20 days	44.00 a	19.10 a	24.86 a	0.84 a
C.V.(%)**	4.24	23.24	13.27	8.59
General mean	43.40	18.70	23.49	0.78

*The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

** Coefficient of variation

On Table 2 we can verify that there is no difference among the sulfur doses for the variables number of leaves per plant and shoot dry weight. For chlorophyll level only the dose of 250 mL classified as inferior, while for plant height the doses of 0 and 500 ml showed the best means. It is highlighted that for chlorophyll level there is interaction between sulfur doses and soybean cultivar, as found by Moreira and Moraes [17], a factor not investigated in the present work. The larger difference observed among the sulfur doses for the variable plant height was an increase of 12%, value similar to the one found by Getachew et al. [18] that verified a response of 16% in plant height when applied 30 kg ha^{-1} of sulfur via soil.

Table 2 - Means of the variables evaluated at full flowering: Chlorophyll level (CLO), number of leaves per plant (NLP), shoot dry weight (SDW) and plant height (PH) on soybean in function of sulfur doses applied.

Doses applied (mL ha^{-1})	CLO (un. ClorofiloG®)	NLP	SDW (kg)	PH (m)
0	43.58 a*	19.88 a	23.37 a	0.82 a
250	41.92 b	17.38 a	22.48 a	0.73 b
500	44.17 a	19.58 a	24.15 a	0.80 a
750	43.17 a	18.25 a	24.63 a	0.77 b
1000	44.17 a	18.38 a	22.85 a	0.77 b

*The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

Ding et al. [5] showed the importance of sulfur in plant metabolism, using tobacco as plant model. The authors stated increases larger than 18% in dry mass when genetically transformed plants for super expression of sulfate transporter, identified in soybean, were grew in environments deficient in the element (pot). Besides that, the chlorophyll level at the leaves and the grain weight were significant better in such plants. So, the experimentation directly on field, as done in this work, without the possibility to rigorous environmental control, impairs the detection of nutrient effects.

Corroborating the results found here, Tiecher et al. [19] also did not verify significant increases in shoot dry mass of the crops as soybean, sorghum, sunflower, millet and mammon with the application of similar sulfur doses. This author used increases doses of sulfur based fertilizers in a soil with low values of the element. On the other hand, Pereira et al. [20] noted increases in stem diameter, plant height and shoot dry mass on soybean with the application of 50 kg ha^{-1} of sulfur. The authors stated increases of 8.42% in stem diameter, 5.78% in plant height and 21.61% in shoot dry mass, in relation to plants without sulfur application. Vitti et al. [21] also stated increases in vegetative growth with sulfur application, regardless the method of application and the nature of de sulfur source.

In an extensive work evaluating the effect of sulfur applied at the soil on soybean for two years, two growth season inside each year and fifteen environments Divito et al. [7] stated interaction between sulfur doses and grain yield. Such authors pointed out relation between chlorophyll level and sulfur concentration on leaves, besides relation between chlorophyll level and N:S ratio. This ratio on leaves was highly correlated with grain yield, more than the absolute sulfur concentration. Getachew et al. [18] also verified relation between sulfur supply on soybean and nitrogen metabolism when they verified responses on nodulation characteristics and grain yield after sulfur provision. Unlike the present work, such authors found increases in shoot dry mass of 29% with sulfur application of 30 kg ha^{-1}.

For the individual effects compounding the grain yield, as the number of pods, number of grain per pod and thousand grain weight, and also for grain yield itself, did not verify significant differences in function of times of sulfur application (Table 3). There were significant effects for the interaction between doses of sulfur and times of application (stages) for thousand grain weight and grain yield.

Table 3 - Means of the variables evaluated at the harvest: number of pods (NP), number of grains per pod (NGP), thousand grain weight (TGW) and grain yield (GY) on soybean in function of times of sulfur application

Times of application	**NP**	**NGP**	**TGW (g)**	**GY (Kg ha^{-1})**
V2	35.31 a*	2.28 a	187.43 a	3440.01 a
V2+10 days	36.48 a	2.25 a	185.29 a	3532.03 a
V2+20 days	37.99 a	2.27 a	182.74 a	3612.90 a
C.V.(%)**	23.70	3.94	5.46	9.32
General mean	36.57	2.27	185.15	3528.31

* The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

** Coefficient of variation

There were no differences in function of the sulfur doses applied on the yield components as the number of pods, number of grains per pod, thousand grain weight and grain yield (Table 4). Richart et al. (2006) evaluated three doses of elemental sulfur (0, 30 and 60 kg ha^{-1}), applied at the soil, and also did not find responses on grain yield and grain mass. Meanwhile, Moreira and Moraes [17] found average responses of eight soybean cultivars, from tropical and sub-tropical origins, of 80.8% on grain yield and 64.7% on the number of pod per plant investigating the sulfur supply via soil. Such authors stated that the variable number of grains per pod is strongly genetically controlled and the nutritional status of the plants has little effect about it, a fact also observed on the present work.

In spite of there was no difference in function of sulfur doses applied for the yield components as number of pods, number of grains per pod, thousand grain weight and grain yield, the highest yield were obtained with the dose of 1,000 mL ha^{-1}. Chandra and Pandey [4] studied several sulfur concentration in nutritive solution on soybean evaluating its effects in many vegetative and reproductive characteristics, as done in this work with foliar application of sulfur in different doses. However, in such controlled situation of nutrient supply and availability of the element in nutritive solution, the authors found effect of sulfur application up to 36% on the number of pods per plant and 33% on the sugar accumulation on leaves, moreover effects on plant height, dry mass of shoot, root and stem, yield and grain quality in terms of protein and carbohydrates.

Table 4 - Means of the variables evaluated at the harvest: number of pods (NP), number of grains per pod (NGP), thousand grain weight (TGW) and grain yield (GY) on soybean in function of sulfur doses applied

Doses applied (mL ha^{-1})	**NP**	**NGP**	**TGW (g)**	**GY (Kg ha^{-1})**
0	38.14 a*	2.24 a	186.92 a	3337.29 a
250	35.68 a	2.24 a	182.86 a	3560.25 a
500	39.63 a	2.27 a	182.86 a	3509.79 a
750	32.31 a	2.32 a	189.48 a	3524.50 a
1000	37.18 a	2.26 a	183.63 a	3709.75 a

*The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

Unfolding the interaction between sulfur doses applied within times of application, for thousand grain weight, verified that on the V2 stage the largest thousand grain weight was obtained with the dose of 750 mL ha^{-1}. Such fact did not happen on the stages V2+10 and V2+20 days (flowering), which effect of doses within time of application did not differ (Table 5).

Table 5 - Unfolding the interaction of sulfur doses within times of application for thousand grain weight

Doses applied (mL ha^{-1})	**V2**	**V2+10 days**	**V2+20 days**
0	185.70 b*	185.61 a	189.45 a
250	179.25 b	186.40 a	182.93 a
500	184.70 b	183.54 a	180.35 a
750	206.56 a	182.21 a	179.65 a
1000	180.91 b	188.69 a	181.30 a

*The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

On the interaction unfolding of sulfur doses within times of application, for grain yield, verified that on the V2 stage the largest yield were obtained with doses larger than 500 mL ha^{-1} (Table 6). We adjusted a regression model for this case, the only that presented a good fit and, thus, justify to be discussed. For sulfur doses within V2 stage the quadratic model with r^2 of 0.86 pointed out the dose of 731 mL ha^{-1} as the dose of maximum grain yield of 3,690 kg ha^{-1}. For the stage V2+10 days only the dose of 500 mL ha^{-1} showed poor. But, for the stage V2+20 days (flowering) there were not differences on grain yield among the sulfur doses applied.

Table 6 - Unfolding the interaction of sulfur doses within times of application for grain yield

Doses applied (mL ha^{-1})	**V2**	**V2+10 days**	**V2+20 days**
0	2926.88 b*	3542.50 a	3542.50 a
250	3315.63 b	3787.50 a	3577.63 a
500	3784.38 a	2867.50 b	3877.50 a
750	3523.31 a	3668.31 a	3381.88 a
1000	3649.88 a	3794.38 a	3685.00 a

* The means followed by the same letter at the columns did not differ at the level of 5% probability by the Scott-Knott test

Corroborating the results found here, Neto et al. [22] also did not find effect of sulfur application on soybean for thousand grain weight, but found it for grain yield. Such authors evaluated two sulfur doses (0.5 and 1 kg ha^{-1}) and two times of application (R1 and R5.1) finding significance for the contrast between sulfur application and its lacking. Other important result presented by them was that only one sulfur application does not differ on grain yield of two applications, regardless the time and dose applied. When comparing only the effect of sulfur doses the authors found the best performance with 0.5 kg ha^{-1}.

In turn, Filho [23] stated sulfur fertilization did not give considerable gains in soybean grain yield. The author still stated there were not increases in sulfur levels on leaves or on the grains, with the same exportation of the nutrient in function of the treatments. However, the author stated that the soybean was sowed under oat straw, a crop that produce high amount of dry mass, and this can be a factor to explain the absence of response to sulfur application. So, it is possible that the sulfur quantity available due to mineralization of the straw had fulfilled the crop demand.

Stipp and Casarin [24] declared that an efficient fertilization should always take account the diagnostic of soil fertility and the plant nutrition, being this a judicious process, which should consider the physical, chemical and foliar analysis, that will indicate the real sulfur absorption by the plants. Among the alternatives to supply sulfur to the plants, the foliar application can be used as a soil complement, due to the big amounts of sulfur demanded by the crops and its importance on the physiology and metabolism. So, the corrections of sulfur deficiency by foliar fertilizers during the development of the soybean provide better nutritive conditions for the plant [25].

4 Conclusions

The time of foliar sulfur application on soybean influenced the shoot dry weight and the plant height, being the V2+20 days the stage most recommended. The responses to the different sulfur doses varied according to the results of chlorophyll level and the plant height. For grain yield there was interaction between time of application and sulfur dose, being the dose of 731 mL ha^{-1} applied at V2 stage the one of maximum yield. The yield components, number of pod per plant and number of grains per pod, were not affected by the time of application or sulfur dose applied. For thousand grain weight, the responses to the different doses varied according to the time of application.

EFICIÊNCIA DO FORNECIMENTO DE ENXOFRE FOLIAR EM DIFERENTES ESTÁDIOS DE DESENVOLVIMENTO DA SOJA

RESUMO: A soja é uma cultura de destaque mundial, cujos grãos têm inúmeros usos na alimentação humana e animal. Dentre os nutrientes essenciais o enxofre é um macronutriente constituinte de proteínas e aminoácidos sendo requerido em quantidade considerável pelas leguminosas. O objetivo do trabalho foi determinar o efeito da utilização de enxofre, via aplicação foliar em diferentes estádios de desenvolvimento da soja, sobre características de desenvolvimento vegetativo e componentes de produtividade. O experimento foi conduzido a campo sob delineamento experimental em blocos casualizados com quatro repetições, em esquema fatorial 3x5 (3 épocas de aplicação via foliar e 5 doses de enxofre). A cultivar de soja TMG 132RR foi utilizada nessa investigação com as doses de 0, 250, 500, 750 ou 1000 mL ha^{-1} de enxofre aplicadas nos estádios V2, V2+10 ou V2+20 dias. A época de aplicação de enxofre foliar influenciou a produção de matéria seca da parte aérea e a altura de plantas, sendo o estádio V2+20 dias a época mais recomendada. A resposta às diferentes doses de enxofre variou com base nos resultados do teor de clorofila foliar e a altura de plantas. Para a produtividade de grãos houve interação entre época de aplicação de enxofre e dose, sendo a dose de 731 mL ha^{-1} aplicada em V2 a de máxima produtividade de3690 Kg ha^{-1}. Os componentes de rendimento número de vagens por planta e número de grãos por vagem não foram afetados pelas épocas ou doses de aplicação de enxofre. Para peso de mil grãos as respostas às diferentes doses variaram conforme a época de aplicação.

Palavras-chave: *Glycine max.* Nutrição mineral. Tratos culturais. Componentes de produtividade.

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