

Dry Mass Increment, Foliar Nutrientes and Soybean Yield as Affected by Aminoacid Application

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Received: August 20, 2019

Accepted: October 11, 2019

Online Published: November 15, 2019

doi:10.5539/jas.v11n18p230

URL: <https://doi.org/10.5539/jas.v11n18p230>

Abstract

Due to the importance of soybeans worldwide, there is a constant search for products or management systems that aim to increase the productivity of this crop. In this sense, some products that have amino acids in their composition have been used, however, there is still a lack of studies that aim to show the isolated effect of amino acids on growth parameters. Therefore, the present study aimed to evaluate the effect of the application of amino acids in the treatment of seeds and of the leaf in the soybean crop. Experiments were carried out in a greenhouse and in the field with the application of glutamate, phenylalanine, cysteine, glycine as a seed treatment (ST) and also as foliar application (FA) at the V₄ growth stage. The dry mass accumulation of root, stem, leaves, total and yield of a soybean crop were evaluated. In addition, leaf element concentration in leaves was also evaluated. The use of phenylalanine in ST promoted the best results on stem mass, leaves, pods and total dry mass, with an increase of up to 152%, as compared to control. This same treatment led to higher productivity, with a 46% increase in relation to the control. In relation to foliar concentration of elements, the most effective application is the one that is carried out in both modes of application (ST and FA), mainly with glutamate and glycine. Therefore, the application of amino acids, especially in seed treatment promotes the greater accumulation of dry mass and productivity in soybean plants.

Keywords: glutamate, cysteine, phenylalanine, glycine, seed yield

1. Introduction

Amino acids are organic molecules formed by carbon, hydrogen, oxygen and nitrogen, and all of them have a carboxyl group (COOH) and an amine group (NH₂) attached to a carbon atom which has attached another hydrogen atom and a (R) radical. The (R) radical represents an organic radical, different in each amino acid molecule found in living matter. Each variation in the amino acid number or sequence produces a different protein and thus, there is a wide variety of proteins. In recent years, a number of amino acid transporters have been discovered in plants, among them LHT1 (Lysine Histidine Transporter 1), AAP1 and AAP5 (Amino Acid Permease 1 and 5). These are present mainly in the roots, but according to Weiland et al. (2015), they can also be found in leaves. This information shows that plants can absorb soil amino acids, as already confirmed by Hirner et al. (2006) and Lee et al. (2007), and indicate that the absorption could possibly happen by the leaf.

In contrast, a large volume of work has shown the effect of amino acids when applied to plants. These can be used as stress-reducing agents, nitrogen source and hormone precursors (Zhao, 2010; DeLille et al., 2011; Maeda & Dudareva, 2012). In addition, more recently amino acids have been proposed as plant flags after the identification of glutamate receptors (GRLs), but which are also capable of binding to other amino acids. When these receptors are activated, several physiological processes are triggered, such as nitrogen uptake regulation (Miller et al., 2007), root development (Walch-Liu & Forde, 2007; Weiland et al., 2015) and antioxidant metabolism (Hildebrandt et al., 2015, Weiland et al., 2015).

These effects together can provide greater accumulation of dry matter mass and changes in the concentration of nutrients in the leaves, which can have repercussions on productivity increases of the soybean crop.

Germination of lupine seeds *in vitro* in a solution containing 35 mM asparagine showed an increase in starch content and a reduction in the concentration of soluble sugars (Borek et al., 2013). However, studies exploring these effects in soybeans are scarce. In addition, most studies have used mixtures of amino acids or formulated products without the proper characterization of amino acids, which makes it difficult to understand the effect that each single amino acid can cause in plants.

The objective of the present work was, therefore, to evaluate the effect of the application of glutamate, cysteine, phenylalanine and glycine (via seeds, leaves or both modes of application) on the accumulation of dry matter mass and nutrients, and on soybean seed yield.

2. Method

2.1 Greenhouse Experiment

This experiment was carried out in Piracicaba, SP, Brazil (22°41'S, 47°38'W and 546 m asl.), using pots of 11 dm³ capacity, filled with washed sand and installed in a plant growth greenhouse. Ten soybean seeds (*Glycine max* L. Merrill), cultivated variety NS 7901 RR were sown in each pot and, after emergence, thinning was done, leaving only three plants per pot.

The experiment was carried out using a randomized block design, consisting of the application of amino acids as seed treatment (ST) and/or foliar application (FA) at V₄ stage (four nodes on the main stem, 20 DAS), using eight blocks per treatment (Table 1). Before application of the treatments, all the seeds were treated with fungicide and insecticide [fipronil (250 g L⁻¹) + methyl thiophanate (225 g L⁻¹) + pyraclostrobin (25 g L⁻¹)] at the rate of 1 mL kg⁻¹ of seeds.

Table 1. Concentration of different amino acids, and time of application, where (ST) were the applications performed only on seeds, (FA) foliar application, and both ST and FA

| Amino acids ¹ | Moment of application | | | |
|--------------------------|---|--|--------------|---------------|
| | Only seed treatment (ST) (mg kg ⁻¹ [seeds]) | Only foliar application (FA) at V ₄ (mg ha ⁻¹) | Both ST + FA | |
| Control | 0 | 0 | 0 | 0 |
| Glutamate (Glu) | 12 | 123 | 12 | 123 |
| Cysteine (Cys) | 12 | 123 | 12 | 123 |
| Phenylalanine (Phe) | 3 | 30 | 3 | 30 |
| Glycine (Gly) | 9 | 92 | 9 | 92 |
| Glu+Cys+Phe+Gly | 12+12+3+9 | 123+123+30+92 | 12+12+3+9 | 123+123+30+92 |

Note. ¹ The sources used equivalent to the pure amino acids, with optical isomerism levogyrous (L-amino acid).

The pots were irrigated daily according to the water requirement (400 mL per pot), and a weekly application of nutrient solution proposed by Johnson et al. (1957) was performed during the experiment.

2.2 Field Experiment

This experiment was conducted in Patos de Minas (MG), Brazil (18°34'S, 46°31'W and 815 m asl.), in the 2014/2015 season. Before sowing, soil samples were collected from the area where the experiment was installed. The chemical analysis of the soil is presented in Table 2. As fertilization, 450 kg ha⁻¹ of formulation 08:30:10 + 3.26% of Ca + 4.25% of S + 0.2% of B + 0.2% Zn were used. Before the installation of this experiment, the area was cultivated with maize for silage.

Table 2. Soil analysis (0-20 cm) of the field experiment

| pH | P-Mel | P-rem | K ⁺ | Ca ²⁺ | Mg ²⁺ | Al ³⁺ | H+Al |
|----------|--|-------|----------------|--|------------------|------------------|----------------------------------|
| in water | ----- mg dm ⁻³ ----- | | ----- | ----- cmol _c dm ⁻³ ----- | | ----- | ----- |
| 5.60 | 10.23 | 4.94 | 85.70 | 2.94 | 0.56 | 0.02 | 1.30 |
| SB | t | T | V | m | OM | | |
| ----- | ----- cmol _c dm ⁻³ ----- | | ----- | ----- % ----- | | ----- | ----- dag kg ⁻¹ ----- |
| 3.72 | 3.74 | 5.02 | 74.11 | 0.53 | 3.12 | | |

Note. P-Mel and K⁺: Mehlich-1 extractor; Ca⁺, Mg⁺ and Al⁺: KCl 1 mol L⁻¹ extractor; P-rem: Remaining phosphorus-phosphorus concentration of the equilibrium solution after stirring 1 hour with 10 mmol L⁻¹ CaCl₂ solution containing 60 g of P (1:10).

Weed management was performed using glyphosate (480 g L⁻¹) at the rate of 2 L ha⁻¹ at 14 and 35 days after sowing (DAS). For pest management, the application of acetamipride (100 g L⁻¹) + alpha-cypermethrin (200 g L⁻¹) at the rate of 250 mL ha⁻¹ and chlorfenapyr (240 g L⁻¹) at the rate of 500 mL ha⁻¹ at 84 DAS. Diseases control was performed with pyraclostrobin (133 g L⁻¹) + epoxiconazole (50 g L⁻¹) at the rate of 600 mL ha⁻¹ at 54 DAS and fluxapyroxad (167 g L⁻¹) + pyraclostrobin (333 g L⁻¹), at the rate of 300 mL ha⁻¹ at 70 and 84 DAS.

The experiment was conducted in a randomized block design, consisting of the application of amino acids as ST and/or FA, using eight blocks per treatment (Table 1). Before application of the treatments all the seeds were treated with fungicide and insecticide [fipronil (250 g L⁻¹) + methyl thiophanate (225 g L⁻¹) + pyraclostrobin (25 g L⁻¹)] at the rate of 1 mL kg⁻¹ of seeds.

Soybean seeds (*Glycine max* L. Merrill), cultivated variety NS 7901 RR, were planted with the objective of reaching a harvesting population of 250,000 plants ha⁻¹. Each experimental unit consisted of four lines seven meters long, with 0.45 m between rows, totaling an area of 12.6 m². In each plot 0.5 m was discarded at each end, making the useful area of each plot only the central lines.

Amino acid applications via foliar were performed at V₄ growth stage with a CO₂ propellant sprayer. The bar used contained four fan-type nozzles, being 2.25 m long and with a pressure of 2 bar. For all applications, a volume of 200 L ha⁻¹ syrup was prepared.

2.3 Measurements

2.3.1 Elements Concentrations in Leaves

The element concentrations in leaves at the V₆ stage - six nodes on the main stem, 20 DAS. P, K, Mg, S, Ca, Zn, Mn, Fe and Cu were determined by X-ray fluorescence spectroscopy of dispersive energy (EDXRF). Dried plant material (1 g) was packed into a polyethylene cup of 20 mm internal diameter and covered with 6-μm thick polypropylene film (Mylar®). The samples were irradiated in duplicate for 300s under vacuum using an energy dispersive X-ray fluorescence spectrometer (Shimadzu EDX-720, Shimadzu, Sao Paulo, SP, Brazil). The element content was calculated using the element concentration and dry mass of leaves.

The quantification of the nitrogen concentration in the leaves was performed by the Kjeldahl method, distilling all the ammonium from the digested leaves in a strongly alkaline medium and retaining the ammonia in a solution of boric acid, which was then titrated with HCl (0.01 M).

2.3.2 Dry Matter Mass

Root, stem, leaf and pod dry mass determinations were performed in the V₆ and R₇ (beginning maturity) stages (40 and 100 DAS), in the greenhouse experiment and in the V₆ and R₈ (full maturity) stages (40 and 110 DAS, respectively), in the field experiment. The analysis was performed by inserting three plants per plot in paper bags, and each fragment was stored separately, drying was performed using the standard forced-air oven drying method at constant temperature of 65 °C, to constant mass.

2.3.3 Seed Yield

In the greenhouse experiment the plants were harvested manually considering three plants per replicate. The grains harvested from each plant were weighed on a digital scale with an accuracy of 0.01 grams. The water content of the grains was determined and the productivity was calculated with the water content corrected to 13% (0.13 g g⁻¹). In the greenhouse experiment, the productivity was determined in grams per plant, from three plants per replicate. In the field experiment, the plants were harvested manually considering the two central

rows, discarding the 0.5 m at each end. After this, weights and measurements were obtained as described for the other experiments and the productivity was determined per unit area.

2.3.4 Statistical Analysis

The data obtained in the two experiments were evaluated for normality and homogeneity using the Shapiro-Wilk and Levene tests, respectively ($p < 0.05$).

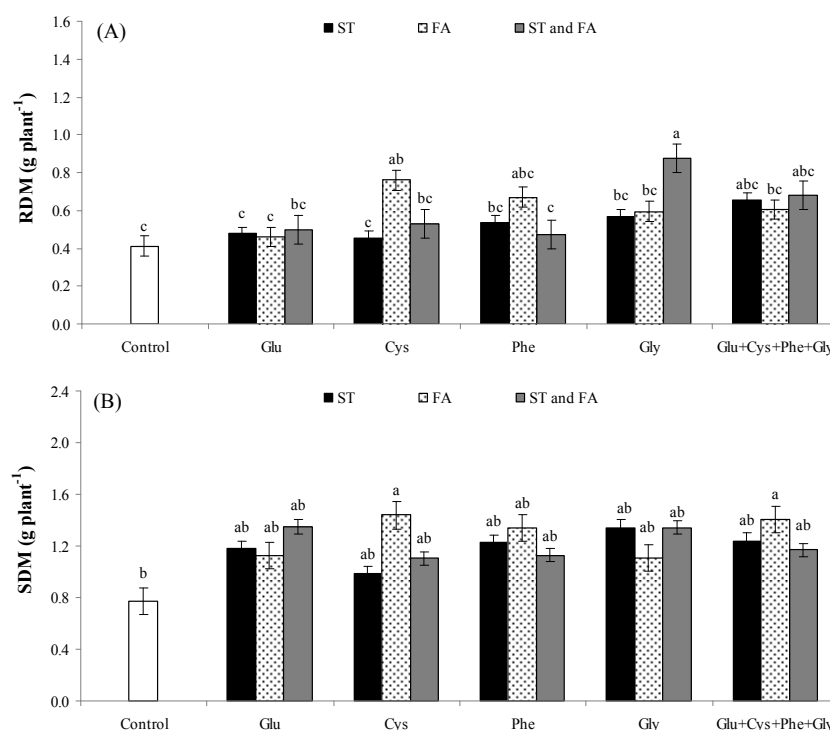
The analysis of variance was performed and, when significant, the Duncan test was applied ($p < 0.05$). For the two experiments, multivariate analyzes were performed through Principal Component analysis. From the analysis, the eigenvalues (values representative of the retained variability for each new component) and eigenvectors (values representative of the location of the points in the graph) were calculated. By means of these data we determined which variables were more representative in the variability of the data, being able, therefore, to generate a two-dimensional chart of the type byplot.

All analyzes were performed using the statistical software SAS 9.3 (SAS Institute, 2011).

3. Results

3.1 Greenhouse Experiment

The application of cysteine in ST and glycine applied in both modes ST and FA presented higher root dry mass of plants at the V_6 stage (Figure 1A). At this same stage, the application of cysteine and set of amino acids via FA raised the stem dry mass in 82% (Figure 1B). Amino acid application had no effect on leaf dry mass and total dry mass (Figures 1C and 1D).



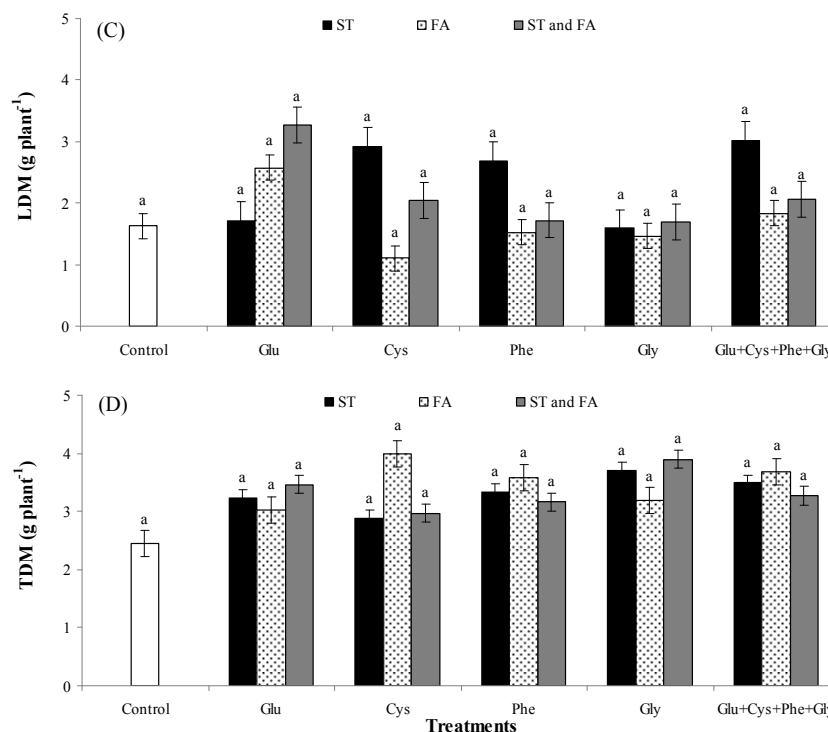
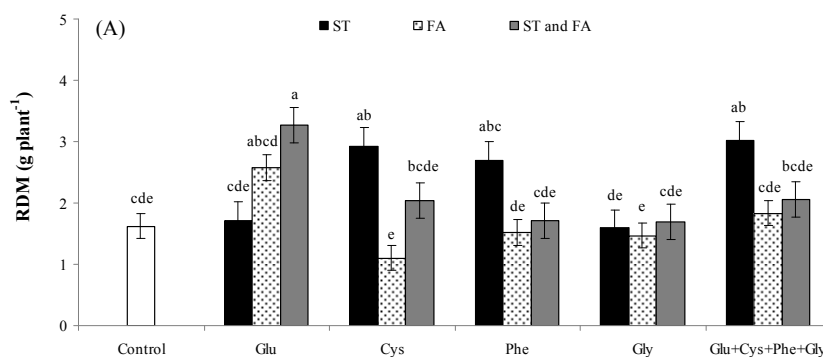


Figure 1. Effects of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the root dry mass (RDM, A), stem dry mass (SDM, B), leaf dry mass (LDM, C) and total dry mass (TDM, D) at V₆ stage. Greenhouse experiment. Season 2014/2015. Means followed by the same letters do not differ significantly from each other, using the Duncan test ($p > 0.05$)

At the R₇ stage, the use of glutamate applied in both modes increased the root and stem dry mass (Figures 2A and 2B). On the other hand, the application of the set of amino acids raised the leaf and total dry mass (Figures 2C and 2D). There were no significant effects of amino acid application on plant productivity compared to control. Only a difference between the treatments with amino acids was observed, in which the use cysteine as ST increased the soybean seed yield by 30% in relation to this same treatment as FA (Figure 2E).

The analysis of main components (PC), indicated two variables with the greatest representativeness (Figure 3), seed yield as PC₁ and total dry mass at R₇ as PC₂.

Positive correlations were found between seed yield and TDM, LDM, RDM and SDM at R₇ stage, knowing that the increase of these variables promotes the increase in seed yield. According to the analysis, the use of phenylalanine and all these amino acids in association with ST and glutamate applied in both modes increased the seed yield.



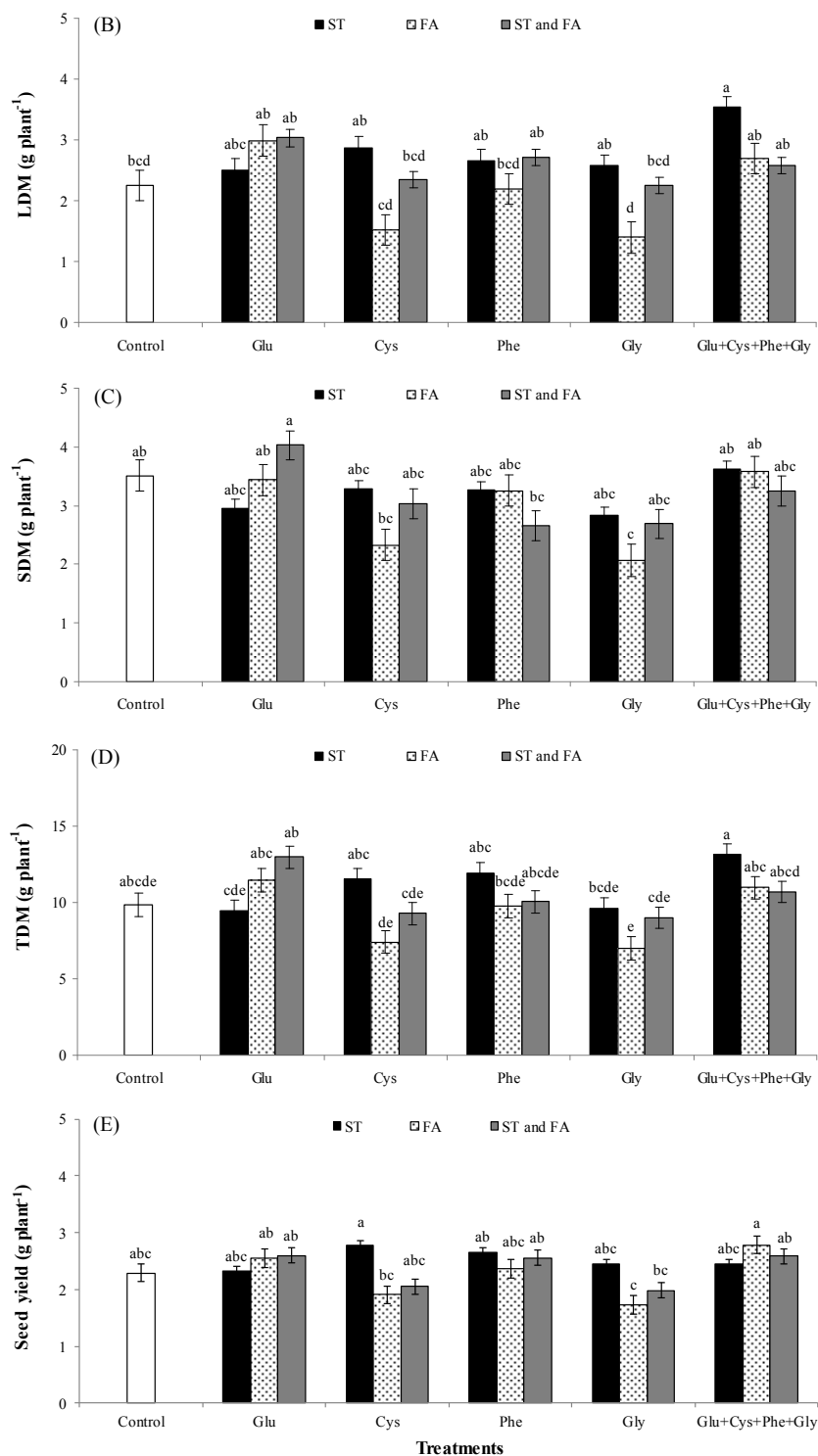


Figure 2. Effects of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the root dry mass (RDM, A), leaf dry mass (LDM, B), stem dry mass (SDM, C) and total dry mass (TDM, D), at R₇ stage and soybean seed yield (E). Greenhouse experiment. Season 2014/2015. Means followed by the same letters do not differ significantly from each other, using the Duncan ($p > 0.05$)

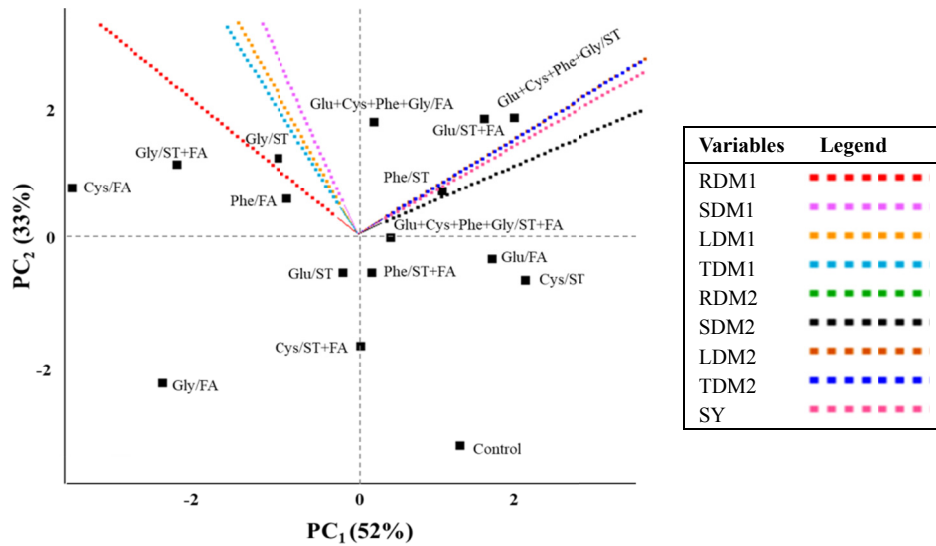
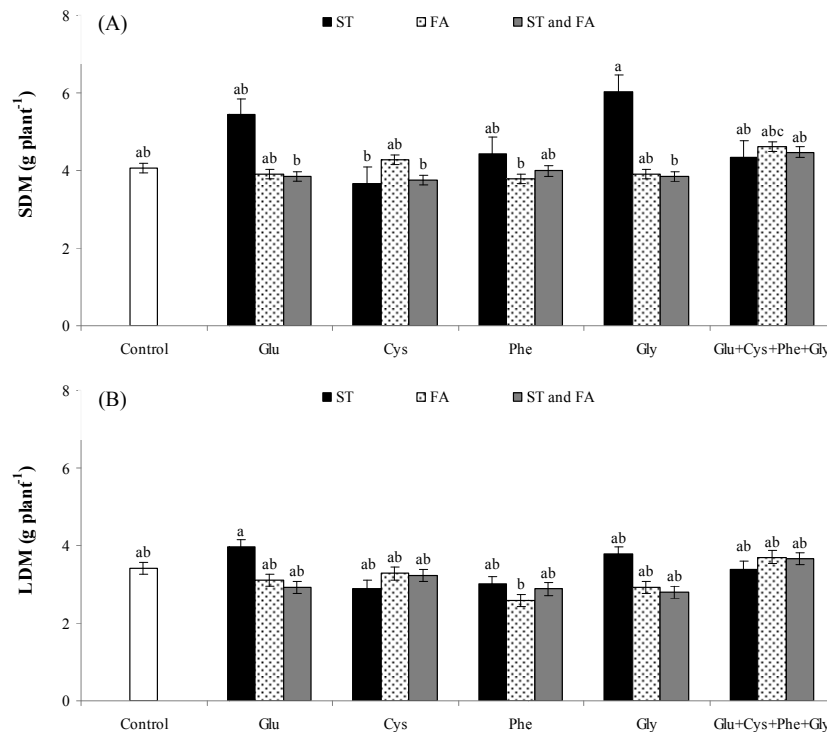


Figure 3. Biplot obtained from the analysis of main components of the results of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the soybean root dry mass at V₆-six nodes on the main stem, 20 DAS (RDM1) and at R₇ stage-beginning maturity, 100 DAS (RDM2), stem dry mass at V₆(SDM1)) and at R₇ stage (SDM2), leaf dry mass at V₆(LDM1) and at R₇ stage (LDM2), total dry mass at V₆(TDM1) and at R₇ stage (TDM2) and soybean seed yield (SY). Greenhouse experiment, Season 2014/2015

3.2 Field Experiment

At the V₆ stage, the use of glycine as ST promoted the increase of stem and total dry mass, representing 48 and 32% increase, respectively, in comparison to the control (Figures 4A and 4C). The leaf dry mass increased with the use of glutamate as ST (Figure 4B).



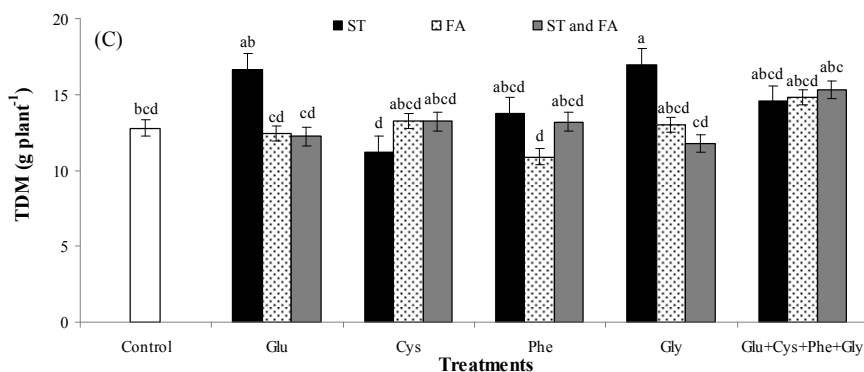
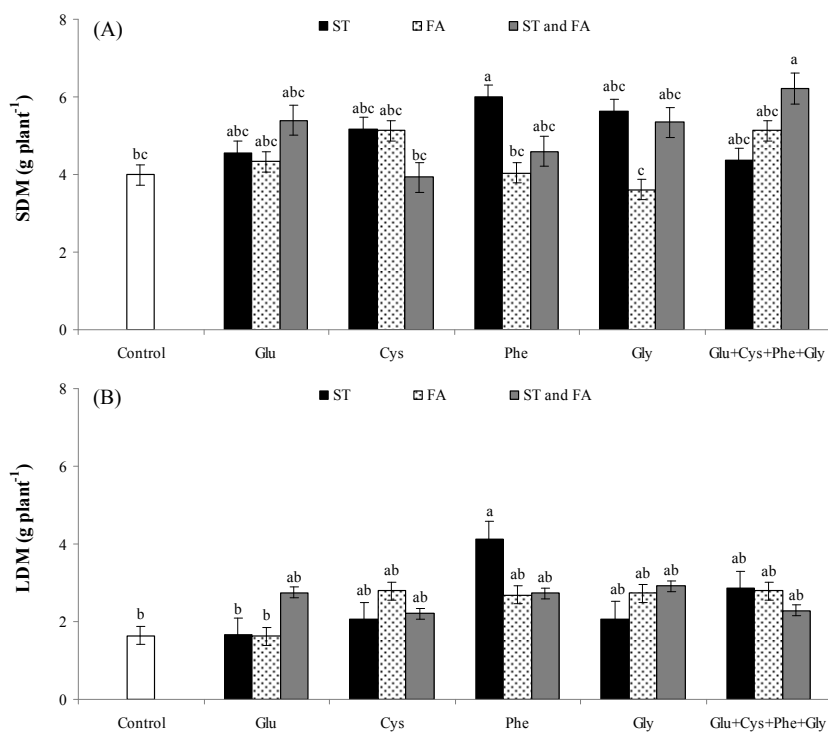


Figure 4. Effects of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the stem dry mass (SDM, A), leaf dry mass (LDM, B) and total dry mass (TDM, C) at V₆ stage. Field experiment, Season 2014/2015. Means followed by the same letters do not differ significantly from each other, using the Duncan test ($p > 0.05$)

On the other hand, at the R₈ stage, the use of phenylalanine as ST provided the best results for stem, leaf, pod and the total dry mass, representing increases of up to 152% compared to the control (Figures 5A, 5B, 5C and 5D). These increases may have favored the higher seed yield of the plants submitted to this treatment, with had a seed yield 46% higher than the control (Figure 5E).



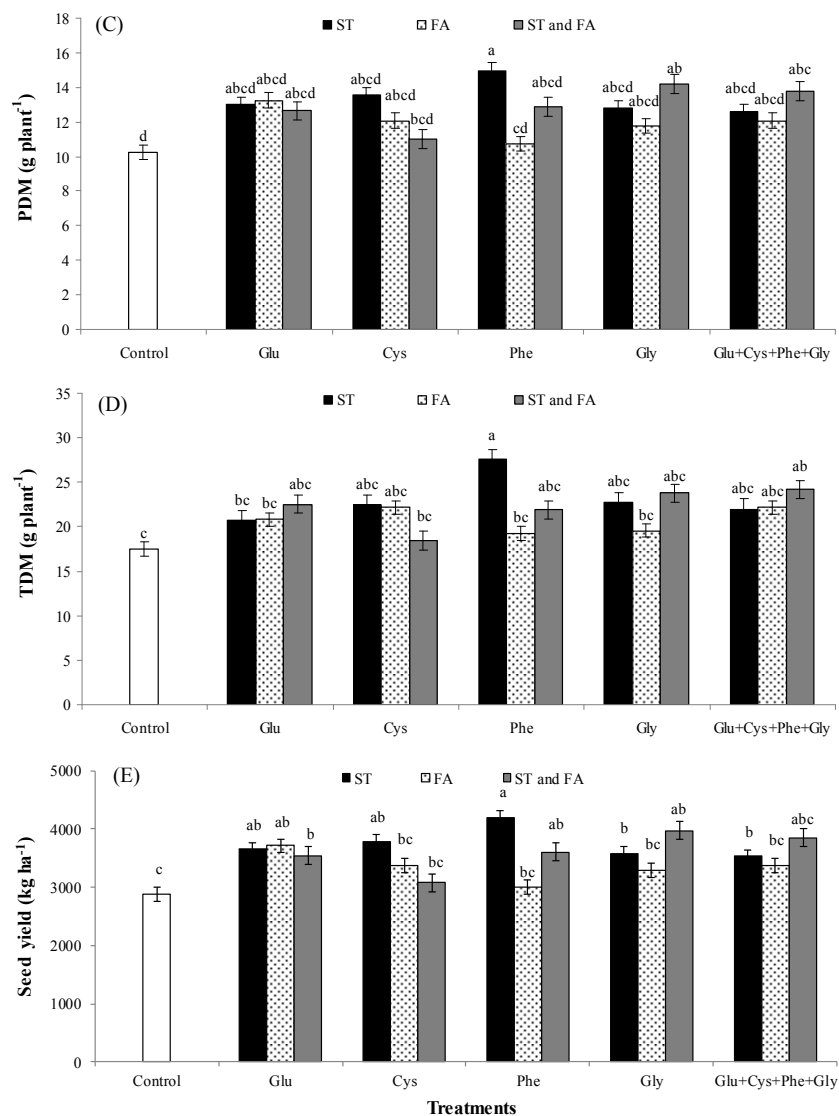


Figure 5. Effects of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the stem dry mass (SDM, A), leaf dry mass (LDM, B), pod dry mass (PDM, C) and total dry mass (TDM, D) at R₈ stage, and soybean seed yield (E). Field experiment, Season 2014/2015. Means followed by the same letters do not differ significantly from each other, using the Duncan test (p > 0.05)

The variables dry mass and seed yield were more important in the analyzes and were represented by PC1 and PC2 (Figure 6). The seed yield showed a higher correlation with total, stem and leaf dry mass, and the treatment with phenylalanine was correlated with the increase in seed yield.

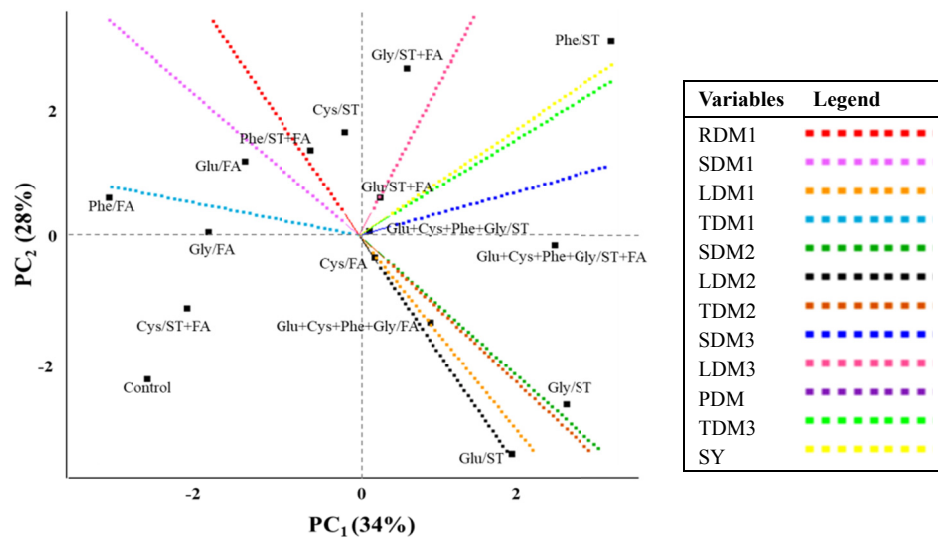


Figure 6. Biplot obtained from the analysis of main components of the results of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the soybean stem dry mass at V₆-six nodes on the main stem, 20 DAS (SDM1) and at R₈ stage-full maturity, 110 DAS (SDM2), leaf dry mass at V₆ (LDM1) and at R₈ stage (LDM2), total dry mass at V₆ (TDM1) and at R₈ stage (TDM2), pod dry mass at R₈ stage (PDM2) and soybean seed yield (SY). Field experiment, Season 2014/2015

Determination of leaf element accumulation at V₆ stage shows a better effect of amino acids when applied in both modes ST and FA. The application of glutamate in these two modes of application increased the contents of K, Mn, Fe, Cu and Zn in leaves, when compared to the control (Table 3).

The use of glycine increased concentration of P and S. For all treatments the total nitrogen in leaves increased in relation to the control. On the other hand, the foliar application of glycine reduced the concentration of several elements in leaves, such as P, K, S and Cu.

Table 3. Effects of use of glutamate (Glu), cysteine (Cys), phenylalanine (Phe), glycine (Gly) and all these amino acids in association (Glu + Cys + Phe + Gly), as function of seed treatment (ST) and foliar application (FA), on the element concentration in leaves at V₆ stage. Field experiment, Season 2014/2015.

| Treatments | Moment of application | Element | | | | | | | | | |
|---------------------|-----------------------|-----------------------|------------|-----------|---------|------------|------------------------|----------|-----------|-------------|------------|
| | | N | P | K | Mg | S | Ca | Mn | Fe | Cu | Zn |
| | | g kg ⁻¹ DM | | | | | mg kg ⁻¹ DM | | | | |
| Control | - | 32.0 b | 2.7 abc | 15.3 abc | 2.6 bc | 1.77 abc | 8.9 ab | 94.3 ab | 240.3 abc | 45.6 ab | 33.4 abc |
| Glutamate (Glu) | | 41.2 a | 2.4 abcdef | 14.9 abcd | 3.0 a | 1.54 de | 8.7 abc | 88.9 ab | 190.1 c | 38.7 cdef | 30.0 abcde |
| Cysteine (Cys) | | 41.2 a | 2.5 abcde | 14.1 cd | 2.7 bc | 1.60 bcde | 7.7 d | 76.1 b | 150.7 c | 38.3 def | 27.7 cde |
| Phenylalanine (Phe) | ST | 40.8 a | 2.3 cdef | 14.6 abcd | 2.7 bc | 1.72 abcde | 8.5 abcd | 100.0 ab | 192.6 bc | 44.3 abcd | 34.6 ab |
| Glycine (Gly) | | 39.5 a | 2.7 abcd | 14.6 abcd | 2.8 abc | 1.74 abcd | 8.1 bcd | 81.6 ab | 213.7 abc | 36.3 ef | 29.4 bcde |
| Glu+Cys+Phe+Gly | | 41.9 a | 2.5 abcde | 14.6 abcd | 2.7 bc | 1.68 abcde | 8.2 bcd | 88.8 ab | 195.8 bc | 39.7 bcd | 25.4 e |
| Glu | | 40.1 a | 2.7 ab | 15.2 abc | 2.9 ab | 1.69 abcde | 9.0 ab | 95.1 ab | 184.2 c | 38.2 def | 32.3 abcd |
| Cys | | 41.0 a | 2.3 cdef | 15.2 abc | 2.5 c | 1.55 cde | 9.2 a | 102.5 a | 236.2 abc | 42.3 abcde | 31.4 abcd |
| Phe | FA | 42.3 a | 2.4 abcdef | 14.1 cd | 2.5 c | 1.49 e | 7.8 cd | 81.7 ab | 178.6 c | 41.6 abcdef | 31.5 abcd |
| Gly | | 40.2 a | 2.1 f | 13.8 d | 2.7 bc | 1.50 e | 8.8 ab | 82.3 ab | 291.2 ab | 35.7 f | 27.2 de |
| Glu+Cys+Phe+Gly | | 38.7 a | 2.2 ef | 14.1 cd | 2.7 abc | 1.61 abcde | 8.6 abcd | 92.5 ab | 216.9 abc | 35.8 f | 26.6 de |
| Glu | | 42.1 a | 2.5 abcde | 15.7 a | 2.8 abc | 1.66 abcde | 8.6 abcd | 104.0 a | 309.2 a | 47.9 a | 35.5 a |
| Cys | | 42.2 a | 2.5 abcde | 15.5 ab | 2.8 abc | 1.79 ab | 8.9 ab | 86.0 ab | 215.9 abc | 45.0 abc | 30.4 abcde |
| Phe | Both ST + FA | 42.1 a | 2.6 abcd | 14.7 abcd | 2.6 bc | 1.67 abcde | 9.2 a | 89.5 ab | 220.6 abc | 40.3 bcdef | 32.3 abcd |
| Gly | | 41.7 a | 2.7 a | 15.3 abc | 2.7 bc | 1.8 a | 8.1 bcd | 92.3 ab | 227.4 abc | 41.9 abcdef | 30.3 abcde |
| Glu+Cys+Phe+Gly | | 45.2 a | 2.5 abcde | 14.4 bcd | 2.5 c | 1.60 bcde | 8.6 abcd | 103.2 a | 178.73 c | 42.1 abcdef | 34.7 ab |

Note. Means followed by the same letters in the column do not differ significantly from each other, using the Duncan test at 5% significance.

4. Discussion

Different responses regarding the application of amino acids in plants were observed in the two experiments. The main effect of these molecules is the signaling of plants (Price et al., 2012; Forde & Roberts, 2014; Santi et al., 2017), and this characteristic is influenced according to the need of the plant at a given moment. Therefore, different environmental conditions may affect different physiological parameters of plants. In the greenhouse experiment the use of cysteine in ST and glycine in both modes ST + FA increased the root dry mass (Fig. 1A). Nagao et al. (2005) observed that the foliar application of proline + lysine at the rates of 80 and 40 mg L⁻¹ resulted in the addition of the total fresh mass of *Lolium multiflorum*. The benefits of increased dry matter mass on seed yield were observed in this experiment, where it was shown that the accumulation of dry matter mass showed a high and positive correlation with seed yield (Figure 3). The set of amino acids were more effective when applied in both ST + FA modes, which increased LDM and TDM at R₇ stage (Figure 2).

The set of amino acids was more effective when applied in both ST + FA modes, which increased LDM and TDM at the R₇ stage (Figure 2).

Glycine, cysteine and glutamate proteins act on plant signaling pathways and can increase oxidative and nitrogen metabolism (Teixeira et al., 2017; Teixeira et al., 2018). However, the various benefits provided by these amino acids were not sufficient to increase the final yield of the plants in the greenhouse experiment.

In the field experiment the best responses in the dry mass variables occurred with the application of amino acids in the seed treatment. Glycine, glutamate and phenylalanine were the most effective amino acids. Glutamate increased LDM at the V₆ stage (Figure 4B). The increase of leaf area is fundamental for plants as it increases the photosynthetic area, which leads to greater photoassimilate production, which can be used during the grain filling phase, and in this way leading to a productivity increment (Board & Modali, 2005).

The set of amino acids in both modes of application increased the TDM at the V₈ stage, a characteristic that also had repercussions on the seed yield increment. These results corroborate Soares et al. (2016), using applications of different amino acids (glutamate, cysteine, glycine, arginine and methionine, at the rates of 31, 30, 34, 42 and 37 mg kg⁻¹ of seeds, respectively), verified increase in soybean seeds yield cultivated in the field. Sadak et al., (2014) developed experiment using foliar application of amino acid mixtures of the commercial product *Vicia faba* consisting of aspartic acid, serine, glutamate, proline, lysine, methionine, isoleucine, leucine, tyrosine, phenylalanine, histidine and arginine. At the dose of 1500 mg L⁻¹, it promoted an increase in plant dry matter mass, as well as increased carbohydrate, polysaccharide content and plant research. The most significant effect was provided by the application of phenylalanine in ST, which increased SDM, LDM, PDM and TDM.

The higher dry mass production leads to the greater amount of energy available for the grain filling stage, thus leading to increased productivity (Board & Modale, 2005). The phenylalanine also acts on important routes of the secondary metabolism, such as the production of flavonoids and lignins, important for plant resistance (Taiz & Zeiger, 2013, Teixeira et al., 2017). In addition, the phenylalanine can act as activated GRLs (Weiland et al., 2015), which act on signaling for changes in root architecture, protection against stress (Weiland et al., 2015, Teixeira et al., 2017) and carbon balance/photosynthesis (Weiland et al., 2015).

Regarding the concentration of element in the plant, the effect of the amino acids on the nutritional status was better evidenced when the application was performed in both ST + FA modes of application. The use of glutamate increased the concentration of K and micronutrients in the leaves (Table 3). K acts on ATPase activation due to its effect on the load balance. In this way, ATP synthesis is directly linked to the availability of potassium. Therefore, the transport of energy from the source to the drain during grain filling is dependent on potassium, which may explain the observed increase in productivity (Marschner, 2012). Increases in the Mn, Fe, Cu and Zn content are also important because, in addition to other functions, these elements act in the path of oxidative metabolism, forming or activating enzymes that eliminate toxic substances produced under stress conditions. In addition, we have already shown that the application of glutamate reduces lipid peroxidation in soybean plants (Teixeira et al., 2017). The application of Glycine in both ST + FA modes of application favored the accumulation of P and S in the leaves, in this case, P is also an extremely important element for grain filling, since it is part of the ATP formation, thus providing energy for this process. In addition, the entry of inorganic phosphorus (Pi) into the chloroplast stroma is required to exit the trioses, which follow for the synthesis of sucrose, which is then transported to drain organs (Buchanan et al., 2000). Therefore, all these characteristics may explain the increase in seed yield from the application of these two amino acids.

The use of the amino acids glutamate, glycine, cysteine and phenylalanine increases plant dry mass and seed yield, especially when applied as seed treatment, with increments of up to 46% in seed yield with the use of

phenylalanine. In relation to foliar concentration of elements, the most effective application is in both modes of application (ST and FA), mainly with glutamate and glycine.

It is suggested that more works are developed with plants of different cultivars and growth habits, in order to evaluate the effect of amino acids in these situations.

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