



## Plant biostimulant on the production of biomass and yield grain of soybean

### Bioestimulante vegetal e períodos de corte na produção de biomassa e rendimentos de grãos de soja

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**RESUMO:** Com o crescente interesse na busca de recursos alimentícios que possam substituir parcialmente o uso de concentrados na alimentação animal, o cultivo da soja para a produção de biomassa e grãos responde como uma alternativa para o setor pecuário. Sendo assim, o objetivo desta pesquisa foi testar doses de fitoestimulante e períodos de corte para avaliar o ganho de fitomassa e a produção de grãos de soja na região do Agreste Meridional do estado de Pernambuco, Brasil. O experimento foi realizado em condições de campo, no município de Garanhuns, avaliando-se quatro doses do fitoestimulante ( $F_1 = 0,00$ ;  $F_2 = 3,75$ ;  $F_3 = 7,50$  e  $F_4 = 11,25$  mL kg<sup>-1</sup> de sementes) e quatro períodos de corte ( $C_1 =$  sem corte;  $C_2 = 30$  DAE;  $C_3 = 35$  DAE e  $C_4 = 40$  DAE). A massa seca das plantas foi aumentada com o corte mais tardiamente, o incremento nesse período foi verificado com a aplicação das maiores doses de fitoestimulante, contribuindo na elevação na altura, número de nós e biomassa verde. A dose de fitoestimulante de 7,5 mL kg<sup>-1</sup> de semente e o período de corte de 40 dias após a emergência (DAE) são os mais indicados para forragem, por proporcionar maior ganho de biomassa. O corte realizado aos 30 dias associado a dose do fitoestimulante de 4,36 mL kg<sup>-1</sup> de sementes proporciona uma maior produção de grãos de rebrota.

**PALAVRAS-CHAVE:** *Glycine max*, produção vegetal, fitoestimulante.

**ABSTRACT:** With the growing interest in the search for food resources that may partially replace the use of concentrates in animal feed, the cultivation of soybeans for the production of biomass and grains responds as an alternative to the livestock sector. Therefore, the objective of this research was to test doses of phytostimulant and cut-off periods to evaluate the phytomass gain and soybean grains yield in the Agreste Meridional region of the state of Pernambuco, Brazil. The experiment was carried out under field conditions, in the city of Garanhuns, evaluating four doses of phytostimulant ( $F_1 = 0.00$ ,  $F_2 = 3.75$ ,  $F_3 = 7.50$  and  $F_4 = 11.25$  mL kg<sup>-1</sup> of seeds) and four cut-off periods ( $C_1 =$  no cut;  $C_2 = 30$  DAE (days after sowing);  $C_3 = 35$  DAE and  $C_4 = 40$  DAE). The dry mass of the plants was increased when cutting was done later, the increment in this period was verified with the application of the higher doses of phytostimulant, contributing to the elevation in height, number of nodes and green biomass. The phytostimulant dose of 7.5 mL kg<sup>-1</sup> of seed and the cut-off period of 40 days after emergence (DAE) are the most indicated for forage, for providing greater biomass gain. Cutting at 30 DAE associated with the phytostimulant dose of 4.36 mL kg<sup>-1</sup> of seeds provided a higher production of regrowth grains.

**KEYWORDS:** *Glycine max*, plant production, phytostimulant.

## INTRODUÇÃO

The soybean (*Glycine max* L. Merrill) among the cultivated leguminous species in the world is the most important (WIJEWARDANA et al., 2019), as well as an essential source of oil and protein for man and livestock is also a source of raw material to obtain biodiesel (KOBBERG et al., 2011). In Brazil, the South and Midwest regions have been outstanding in the national production, with the states of Mato Grosso, Rio Grande do Sul and Paraná being respectively the largest national producers (CONAB, 2019). Thanks to genetic breeding programs, soybean is currently widely cultivated in Brazil, including in the Northeast region (VIANA et al., 2013). Western of Bahia state, and southern of Piauí and Maranhão states, are consolidated as the largest soybean producers in the Northeast, according to data from the CONAB (2019).

The nutritional quality of soybean contributes to high concentrations of protein mass (35,7%), oil (19,5%) (ASSEFA et al., 2019) and still essential amino acids (KOBBERG et al., 2011). The biomass of leguminous plants has a higher nutrient richness than cereals and during the vegetative growth they present high nutritive value, but as they pass from vegetative to reproductive growth this value decreases markedly (YAO et al., 2018). In addition, legume species have the ability to fix nitrogen in the soil, improving their fertility and being economically viable (RECKLING et al., 2016), standing out as excellent substitutes for nitrogen fertilizers.

In order to increase the productivity of soybeans, new products appear every year to inoculate the seeds and/or to apply them on the leaf and those substances promote the vegetative growth and can act alone or in combination in the increasing of the development of the plants (ZAHOOR, 2017), which can promote biomass gains, contributing to the increase of dry mass. According to Small and Degenhardt (2018) the plant growth regulators have been shown in the literature in various applications to improve germination, establishment, growth and development of crops. The hormones contained in the phytohormones are signaling molecules, naturally present in plants in basically small concentrations, and are responsible for marked effects on plant development (TAIZ; ZEIGER, 2017), influencing or modifying the physiological processes, in order to control the activities related to plant metabolism (ZAHOOR, 2017).

The cutting of plants in agriculture is a way to stimulate the growth and development of the crop, because when removing organs or parts of the plant, the plant organism goes through a process of physiological stress, which changes the rate of hormones. In soybeans, the cut is a strong ally to increase the amount of branching, consequently increasing the amounts of pods produced by plants and decreasing the lodging (TOLEDO et al., 2009). In animal feed, soybeans submitted to cutting can be used in the nutrition of cattle, goats and sheep as forage (CASTRO-MONTOYA et al., 2018). The use of dual purpose soybeans (forage and grains) depends on the influence of the environment conditions so that the grains of the regrowth are of good quality (TANCREDI et al., 2006). The cutting of the plants, 30 cm from the ground, and later use of the regrowth grains is feasible, allowing one more option to the agriculturist: production of hay and grains in the same crop (REZENDE et al., 2003).

With the growing interest in the search for food resources that could partially replace the use of concentrates in animal feed and the lack of studies of cutting and use of phytostimulant, the objective was to test phytostimulant doses and cut-off periods to evaluate the phytomass gain and the production of grains in soybean cultivated in the Agreste Meridional region of Pernambuco, Brazil.

## MATERIAL AND METHODS

The experiment was carried out in field conditions, in the agricultural year of 2012, in the city of Garanhuns, Pernambuco, in the Morada Nova farm, latitude 8°56'36 "south, longitude 36°33'03" west, being at an average altitude of 772 m (EARTH, 2012). The predominant climate in the region is the 'As' type, which is equivalent to a hot and humid climate according to Köeppen classification (MOTA; AGENDES, 1986), with average annual temperature of 20°C and average annual rainfall of 1,038 mm, being the rainiest months from May to June. The relative humidity of the air varies from 75 to 83% (ANDRADE et al., 2008) and the soil classification of the experimental area is Yellow Podzolic (SANTOS, 2011b).

In the experimental area, soil samples were collected before sowing the soybean, to perform soil chemical analysis (Table 1). From the result of the chemical analysis of the soil, the fertilization recommendation was made according to the Cavalcante

(2008). The pH correction for the 6.0 range was through the use of dolomitic limestone, which was applied three months before planting. The soybean cultivar P99R01 of the company Pionner® with Degree of Maturation (GM) 9,0 and cycle of 125-130 days for Homogeneous Environmental Zone (ZAH) V, SubZAH of 5.2, was used for the state of Pernambuco (PIONNER, 2012).

**Table 1.** Chemical soil characteristics of the experimental area.

pH (H <sub>2</sub> O)	P	K <sup>+</sup>	Na <sup>+</sup>	Ca <sup>++</sup>	Mg <sup>+</sup>	Al <sup>+++</sup>	H <sup>+</sup> + Al <sup>+++</sup>	S	CTC	V	m
	mg kg <sup>-1</sup>	-----cmol <sub>c</sub> dm <sup>-3</sup> -----								----- % -----	
4,81	7,53	0,09	0,19	0,1	0,9	0,5	2,15	1,28	3,43	37,39	28,07

To meet nitrogen requirements, 0.2 mL of Rizo-Forte® commercial inoculum from Rizobacter company was used for each kilogram of soybean seed, composed of SEMIA 5079 and SEMIA 5080 strains of the Bradyrhizobium japonicum species, and inoculant application was made on the seeds, two hours before planting and 10 hours after application of phytostimulant. Stimulate® phytostimulant was applied to soybean seeds with the aid of a volumetric pipette 12 hours prior to sowing at the doses of 0, 3.75, 7.50 and 11.35 mL.kg<sup>-1</sup> of seed. The cuts were performed at 5.0 cm from the soil surface at 30, 35 and 40 DAE (days after emergence) and also a treatment of no cut.

Weed control was performed biweekly through manual weeding. During the experiment period there was no need to control pests and diseases. Irrigation was carried out according to the water requirement, according to the precipitation and the daily evapotranspiration of the crop, obtained using the Hagreaves-Samani equation calibrated to Garanhuns-PE micro-region (BORGES JÚNIOR et al., 2012).

The variables analyzed in the cut-off periods were: *Plant height at cut period* (APPC): measured at each cutting period studied, from the soil surface to the end of the main stem; *Number of nodes in the cut-off period*: counting done directly in the working area before the corresponding cut-off period; *Leaf area index*: the total leaf area of the plant (m<sup>2</sup>) per unit of land (m<sup>2</sup>) was calculated for the plant in the cut period; *Green shoot biomass*: obtained by weighing the green sectioned material 5.0 cm from the ground, in the working area, immediately after the cutting period, with the aid of a portable digital scale; *dry shoot biomass*: the green shoot biomass obtained in the

field was conditioned in an air circulation oven at 80 °C, weighed 24 hours later (Nakagawa, 1999). The results were converted into kg ha<sup>-1</sup>.

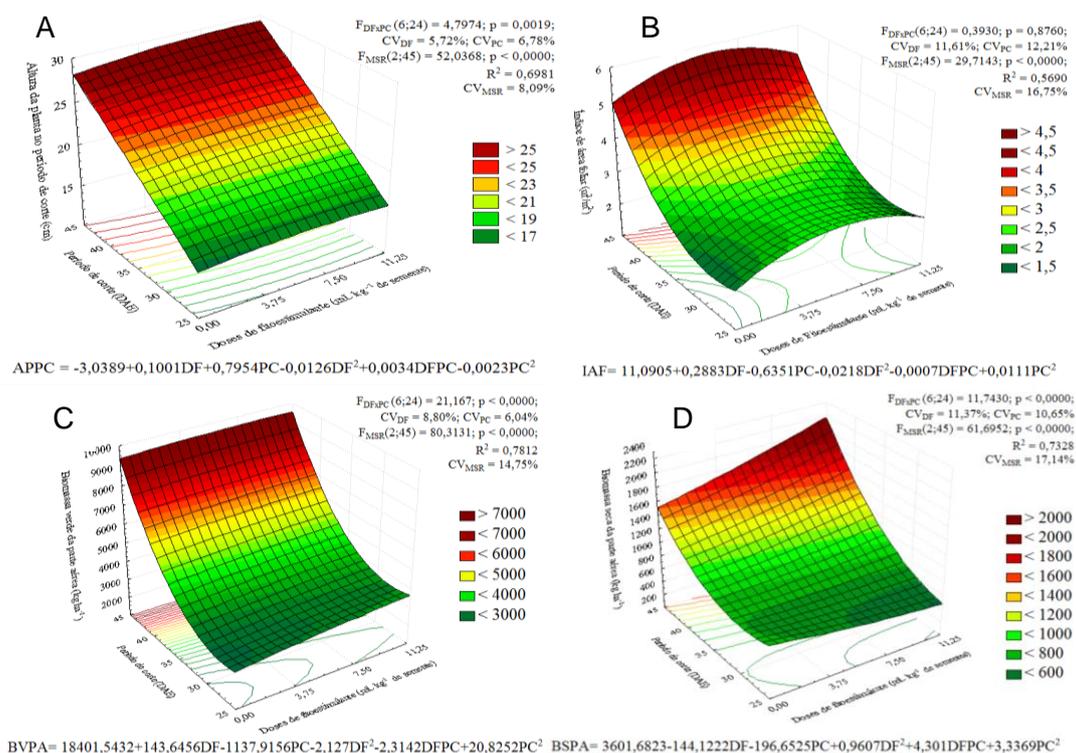
The variables analyzed at harvest were: *Plant height at harvest*: measured from the soil surface to the end of the main stem when the plants were at full maturity (R8); *Height of first pod insertion*: measured from the soil surface to the first pod on the main stem when the plants were at harvest point (R8); *Number of pods per plant* and *Number of nodes at harvest*: counting of pods and nodes performed directly in the usable area before harvest; *Pod* and *grain* productivity: obtained through the average pod and grain yield of the treatments and transformed to kg ha<sup>-1</sup>.

The experimental design was a randomized block design with four replications in plot and subplot plot, where the plots were formed by the phytostimulant doses and the subplots by the cut periods. The sample consisted of 15 random plants in the central square meter (2 m x 0.5 m) of the subplot for each treatment (useful area). Data were submitted to analysis of variance, multiple regression and binomial correlation with the minimum significance level established for null hypothesis ( $p < 0.05$ ). Statistical analyzes were performed with the *software* SISVAR, version 5.3 (FERREIRA, 2010) and STATISTICA, version 8.0 (STAT SOFT, 2007).

## RESULTS AND DISCUSSION

It is observed in Figure 1 that when the cut was anticipated, the height in the plant was 18 cm and in the last cut period (40 days after emergence) reached 26 cm, demonstrating an increase of phytomass with the highest height, before flowering, which occurred at 52 days after sowing. Regarding the application of phytostimulant, the highest dose of 11.25 mL kg<sup>-1</sup> provided a maximum height of 27.99 cm. It is observed that as the cut period and the dose of phytostimulant increase, a proportional increase occurs in the height of the soybean plants at the time of the cut, which is contrary to the information of Buzzello (2010), who states that the hormones (auxins, gibberilins and cytokinin) of the phyto regulators group present good efficiency in reducing plant height. It also contradicts Rademacher (2000), who reports that the use of synthetic phytostimulants reduces the longitudinal growth of the aerial part of the plants, without decreasing grain yield.

Different responses to the application of these products are related to extrinsic and intrinsic factors, as hormones are responsible for remarkable effects on plant development in small concentrations (TAIZ; ZEIGER 2017), especially when different genotypes are evaluated.



**Figure 1.** A- Height of soybean plants, B- Leaf area index, C- Green shoot biomass and D- Dry shoot biomass as a function of phytostimulant (DF) and cut-off periods (PC).

In the state of PiauÍ-Brazil, Lopes (2011) evaluated the agronomic characteristics of the same soybean cultivar without the use of phytostimulant, and verified that the plants cut at 43 days after sowing had a mean height of 61.35 cm and produced 3,425 kg ha<sup>-1</sup> of soybeans grains. Santos et al. (2011a), working with the soybean cultivated in lowland, verified a height of 25.50 cm in the plants that only received the cut at the moment of the harvest yielding productivity of 2,568.00 kg ha<sup>-1</sup>. However, in the present research, the phytomass gain with the increase of the doses provided a gain of green material for cutting and that can be used for making hay, one can still obtain grain production, which is important for agricultural activity.

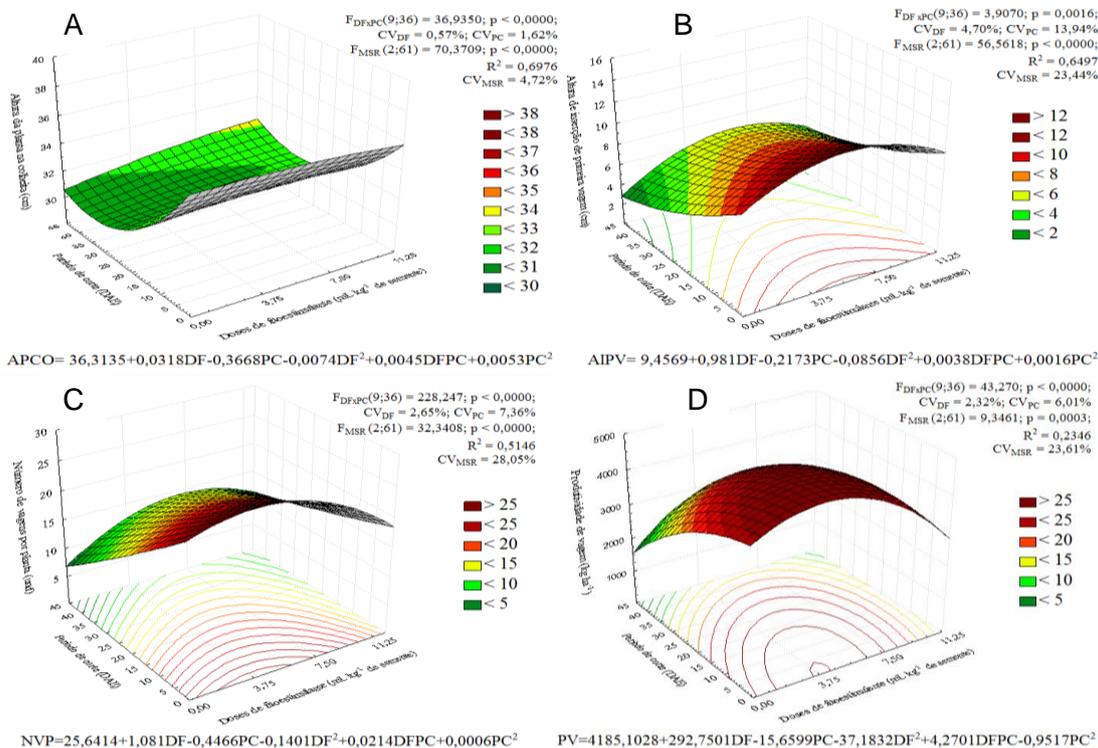
Higher leaf area index was observed in plants that were cut in the last period (Figure 1B), whereas the dose of 7.50 mL kg<sup>-1</sup> of seed provided higher gain with maximum leaf area index of 5.91 m<sup>2</sup> m<sup>-2</sup>. Considering the above, the association of the dose of 7.50 mL kg<sup>-1</sup> of seeds and a cut at 40 DAE will present a good coverage of the area, being this index a great parameter for predicting the biomass productivity. The leaf area index is an important parameter that allows the soybean crop to maximize the use of environmental resources (SEDIYAMA, 2009). It is important to identify the leaf area index of soybean plants to verify the interception of the maximum amount of solar radiation, which is around 95%. For this, it is necessary that the plant presents leaf area index of 3.5 to 4.0 before the R1 stage (BOARD; HARVILLE, 1992), which was found in this research.

The highest values of green biomass of the soybean shoot after cutting were observed in the last cut period (Figure 1C), with a slight increase with the use of phytostimulant. The maximum values obtained at the 40 DAE were similar to those obtained by Santos (1983), who reached 50 DAE values between 4,271 and 10,117 kg ha<sup>-1</sup> of green biomass. The highest values were found by Gris (2006), which obtained values ranging from 19,733 to 30,06 kg ha<sup>-1</sup>, but these cuts were performed from the R<sub>2</sub> reproductive stage of soybean, therefore, after the vegetative phase and at full flowering. Green biomass can be used in fresh form (green or in natural fertilization for livestock) or as silage that can be stored for long periods of time and can be supplied to livestock when pasture or feed availability. The use of phytomoniums has provided positive effects when applied via seeds, with increasing emergence increasing, emergence speed index, biological productivity, vegetative production, seed production and leaf area (MELO et al., 2018). Leaf application to sesame cultivation promoted the increase of fruit number by plants and the concentration of chlorophyll A and B (VIANA et al., 2018), in soybean studies in northern Brazil, the application provided increase in plant height, higher production of root dry mass and increase of root system volume in both seed and leaf application (SANTOS et al., 2017).

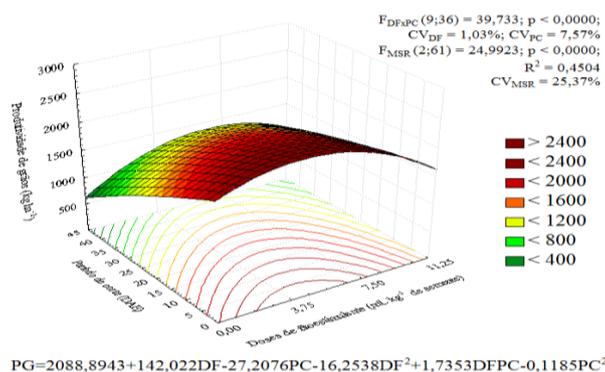
According to the average data of Figure 1D, it was verified that there was an increase of the dry mass of the plants when they were cut later and the increment in that period was increased with the application of the higher doses of phytostimulant, contributing with the previous results, in which showed increase in height, number of nodes and green biomass. Dry biomass is the main component of hay after high loss of

water content. Due to its nutritional characteristics, soybean represents excellent forage potential and if the purpose is biomass and grain production, the use of long cycle cultivars is the best option.

With the later cuts, it was found (Figure 2A) that the heights of the plants at the time of harvest were lower for those cut later than those cut at the beginning of the plant growth, in other words, the earlier cut plants had more time to recover and grow until the time of harvest. It can be seen from the data of Figure 2A that in the initial period of cut, phytostimulant did not provide phytomass gain. However, for the last cut periods, the higher phytostimulant doses established higher plant height gain. The maximum height value of 36.35 cm was provided with the application of 2.15 mL kg<sup>-1</sup> of soybean seeds. After 50 DAS, Santos (1983) observed that the cut soybean increased again up to 86 cm. However, Santos e Vieira (1977), studying the cultivation of soybeans with double purpose, observed that some cultivars presented height after cut at 60 DAS of 35.4, 38.4 and 43.8 cm, respectively, therefore, close to those observed in this study, however, with reduction of 15 days of cycle, since the last cut was performed at 45 DAS.



E



**Figure 2.** A- Plant height at harvest; B- First pod insertion height; C- Number of pods per plant; D- Productivity of pods and E-Grain productivity as a function of phytostimulant (DF) and cut-off periods (PC).

Analyzing the height of insertion of the first pod (Figure 2B), it can be observed that the plants that were cut late presented lower height than those that were cut at the beginning of the vegetative growth, showing that when cutting is at the beginning of the growth it increases the height of insertion of the first pod. The maximum height up to the first pod of 14.85 cm was observed with the application of 5.73 mL of phytostimulant and this is desirable for mechanical harvesting, because according to Sedyama (2009), height above 10 cm avoids harvest losses of soybeans. Plants that did not receive doses of phytostimulant had the first pod insertion height of less than 10 cm. Rezende (2001) verified that the soybean plants that did not receive the cut showed highest height of insertion of the first pod of 13 cm and the plants with the cut at 75 DAE provided the lowest one (7.00 cm). Other authors (SANTOS, 1981; REZENDE; FAVORETTO, 1987; GRIS, 2008; CASTRO-MONTOYA et al., 2018) presented results compatible with those obtained in this study.

According to the data of Figure 2C, there is an increase in the number of pods per plant when they were not cut. The plants submitted to the cut had an average of 22 pods per plant with the application of 7.50 mL kg<sup>-1</sup> of the phytostimulant. The other doses were provided less than 20 pods per plant. Regarding the application of phytostimulant, the equation was quadratic, where the value of 3.85 mL kg<sup>-1</sup> provided a maximum number of pods per plant (28 pods). Biostimulants contribute positively to the following variables: first pod insertion height, plant height, root dry mass and number of branches per plant for soybean cultivation, however for commercial use it is economically unfeasible (FARIA, 2017).

The yield of pods (Figure 2D) presented mean values above 4000 kg ha<sup>-1</sup> for the first three doses of phytostimulant (0.00, 3.75 and 7.50 mL.kg<sup>-1</sup>) that did not receive the cut treatment. The productivity was above of 4000 kg ha<sup>-1</sup> at the doses of 3.75 and 7.50 mL kg<sup>-1</sup> when performing the cuts at the beginning of the vegetative growth indicating the stimulation provided to the soybean plants, since the productivity of the control treatment (without phytostimulant) with the accomplishment of the cuts was below 3,000 kg ha<sup>-1</sup>. The maximum pod yield (4,762.31 kg ha<sup>-1</sup>) can be observed when applying the 3.93 mL kg<sup>-1</sup> of phytostimulant and no cutting (Figure 2D). Campos et al. (2009) studying the application of hormones in soybean found that the height of soybean plants when receiving the dosages increased their height, promoting the vertical growth of plants.

The regrowth of soybean plants provided good pod yield despite treatments showed few nodes and height plant less than 30 cm. Santos (2011a), working with soybean in floodplain in the state of Tocantins-Brazil, observed a high production of pods with harvest height of 25.5 cm in plants without cutting.

The grain yield (Figure 2E) was above 1,900 kg ha<sup>-1</sup> in the three doses tested in the plants that did not receive the cut. The cut plantas presented grain yield below 2,000 kg ha<sup>-1</sup>, but the cuts performed in the initial periods of vegetative growth and associated with the phytostimulant dose of 7.50 mL kg<sup>-1</sup> provided a productivity of up to 1,980 kg ha<sup>-1</sup>. These values demonstrate the efficiency of phytostimulant use in soybean regrowth, as the grain yield of plants that did not receive phytostimulant was less than 1,200 kg ha<sup>-1</sup> in all cut-off periods. The estimated maximum value (2,637 kg ha<sup>-1</sup>) of grain yield was observed when the dose of 4.36 mL kg<sup>-1</sup> was used of seeds.

Rezende (2001) found that yields of soybean grains obtained from cuttings ranged from 977 to 2,058 kg ha<sup>-1</sup>. The differential response to regrowth in previous studies (REZENDE & TAKAHASHI, 1990; REZENDE, 2001) has been observed due to the rupture of apical dominance, thus favoring the presence of a greater or lesser amount of the relation of cytokinin/auxin, which are hormones responsible for stimulating the regrowth of plants. The cut-off period is an inverse relation to grain production, in other words, when the cutting is performed earlier, there is a lower yield of forage, but there is a higher grain yield of the regrowth. If the opposite occurs, when cuts are made later, there is a higher yield of forage, but low yields of grains.

## CONCLUSION

The phytostimulant dose of 7.5 mL kg<sup>-1</sup> of seed and the cut-off period of 40 days after emergence (DAE) are the most indicated for forage, because it provides greater biomass gain.

The cut performed at 30 DAE associated with the phytostimulant dose of 4.36 mL kg<sup>-1</sup> of seeds provides a higher grain yield.

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