

## **Liquid fertilization and conventional fertilization on soil fertility and agronomic and morphophysiological characteristics of *Brachiaria brizantha* cv. Braúna grass**

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**Abstract.** The objective of this study was to evaluate the effect of liquid and conventional fertilization on soil fertility, assessed by regrowth, and on the morphophysiological characteristics of *Brachiaria brizantha* cv. Braúna. The experiment was conducted in a greenhouse at the State University of Southwest Bahia, in Itapetinga-BA, from December 2018 to April 2019. The experimental design adopted was completely randomized, with four replicates, totaling 24 experimental units, in a 2×3 factorial scheme, comprising two types of fertilization (liquid and conventional) and three regrowths, with cutting intervals of 21 days. The results revealed that conventional fertilization promoted an increase in the levels of calcium (Ca) and phosphorus (P) in the soil, which, in turn, positively impacted the sum of bases and base saturation, reaching values of 5.6 cmolc dm<sup>-3</sup> and 54%, respectively, in the first regrowth. The dry mass production of the aerial part was influenced by both regrowth and the type of fertilization. A 27% reduction in dry mass production was observed from the first to the second regrowth, and a 16% reduction from the second to the third. On the other hand, conventional fertilization provided a 20% increase in dry mass production. The morphological variables of the plant were influenced by regrowth, with the exception of the leaf appearance rate (LAR), which did not present significant variations. A reduction in the other morphological variables was observed with each regrowth. Fertilization, in turn, influenced only the length of the flagellum leaf (LFL) and the total length of the tiller (TTL). The interaction between regrowth and fertilization was significant for leaf area, specific leaf area, leaf area index and leaf area ratio. The SPAD index, chlorophyll *b* and total chlorophyll contents were influenced by regrowth, with a progressive increase with each regrowth. Conventional fertilization promoted an increase in the SPAD index, with a value of 35.53, compared to 33.51 for liquid fertilization. The interaction between regrowth and fertilization was significant for the chlorophyll *a:b* ratio and carotenoids. The chlorophyll *a:b* ratio decreased with each regrowth. Conventional fertilization resulted in a 1.04% increase in crude protein content compared to liquid fertilization. Dry matter (DM), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ash contents were influenced by regrowth, with a progressive increase with each regrowth, except for ash, which showed a reduction. Regrowth demonstrated greater influence on the productive and morphophysiological characteristics of braúna grass. Conventional fertilization is recommended to promote improvements in soil fertility and increase the crude protein content of signal grass *Brachiaria brizantha* cv. Braúna.

**Key words:** liquid fertilizer, nutritional composition, plant morphology, regrowth.

## INTRODUCTION

Brazilian livestock farming is largely based on pasture-based cattle raising, a practice that stands out for its economically efficient meat and milk production, giving the country a competitive position in the global market. However, persistently low animal productivity rates pose a significant challenge for the sector.

This low productivity is largely a reflection of the scarcity of forage available throughout the year. This scarcity, in turn, is influenced by a series of interconnected factors, such as low soil fertility, high stocking rates, and neglect of pasture maintenance, particularly with regard to fertilization and soil correction. The combination of these factors triggers a process of pasture degradation, compromising the carrying capacity and quality of the forage available to animals.

In this scenario, intensifying pasture use emerges as a fundamental strategy to ensure the availability of forage in adequate quantity and quality. This intensification entails the adoption of management practices aimed at optimizing forage production, such as fertilization and soil amendment, invasive plant control, and the use of cultivars adapted to local soil and climate conditions (Dutra et al., 2025). Implementing these practices, combined with the adoption of innovative technologies, is essential to reversing pasture degradation and ensuring the sustainability of livestock production.

The intensification of agricultural production essentially consists of implementing technologies aimed at optimizing the results of rural enterprises. These technologies aim to promote significant changes in variables related to forage production, resulting in increased productivity and pasture quality.

The *Brachiaria brizantha* cv. Braúna cultivar has agronomic characteristics that make it a promising option for livestock production. Its dry matter production capacity, which ranges from 8 to 12 tons per hectare per year, combined with its crude protein content, which can reach 12% of the dry matter, makes it a forage crop with high productive and nutritional potential (Matsuda, 2021).

However, the performance of the Braúna cultivar is intrinsically linked to soil fertility, requiring medium- to high-fertility soils to achieve its full potential. A notable characteristic of this cultivar is its vigorous regrowth, which gives it a high recovery capacity after cutting or grazing (Matsuda, 2021). However, the scientific literature still lacks detailed information on the ideal rest period to ensure complete plant recovery. Determining this period is crucial to optimizing pasture management and ensuring the continuous production of high-quality forage.

There are currently several fertilizers for pastures, with granulated (conventional) fertilizers being the best known and most used. However, the difficulty of application and the demand for labor to carry out the application led the market to launch liquid fertilizers, which promise similar results to granulated fertilizers with greater ease of application, lower labor costs and the price of equivalent products.

Granular fertilizers are still the most sought after by livestock farmers, primarily due to their long history of availability and proven effectiveness. Liquid fertilizers, on the other hand, are gaining traction. Despite their short introduction into the market, they have already attracted the attention of users seeking practical application, which is the

main advantage of liquid fertilizer over granulated fertilizer. Furthermore, liquid fertilizers promise faster nutrient delivery to plants by using water as a carrier.

Therefore, the objective of this study was to evaluate the effect of applying liquid fertilizer as a substitute for conventional fertilizer on soil fertility and the structural, physiological, anatomical, and productive characteristics of *Brachiaria brizantha* cv. Braúna.

## MATERIALS AND METHODS

The research with *Brachiaria brizantha* cv. Braúna was carried out in a greenhouse, located at the State University of Southwest Bahia, Juvino Oliveira Campus, Itapetinga, BA (15°38'46" south latitude, 40°15'24", west longitude and with an average altitude of 280 m), during the period from December 2018 to April 2019.

The experiment was conducted in a completely randomized experimental design, in a 2×3 factorial scheme, with two types of fertilization, conventional and liquid, and three regrowths with a defoliation frequency of 21 days, with 4 replicates, totaling 24 experimental units.

The soil used was collected in the 0–20 cm depth layer, classified as Sandy Clay textured soil (according to soil analysis protocol), on the Valeu Boi farm, located in the municipality of Encruzilhada-BA, under the coordinates: latitude 15°31'49" South, longitude 40°54'37" West. The collected soil was broken up and passed through a sieve with a four-millimeter mesh. Material was collected for soil analysis and the pots, with a capacity of 14 L, were filled with 10 kg of dry soil. The results of the soil analysis are presented in Table 1.

**Table 1.** Chemical analysis of soil sample

pH	Mg dm <sup>-3</sup>	Cmol <sub>c</sub> dm <sup>-3</sup> of soil									%		g dm <sup>-3</sup>
(H <sub>2</sub> O)	P	K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Al <sup>3+</sup>	H <sup>+</sup>	S.B	t	T	V	m	M.O	
4.8	6	0.41	1.7	1.2	0.5	5.9	3.3	3.8	9.7	34	13	25	

S.B: Sum of bases; t: effective cation exchange capacity; T: pH cation exchange capacity; V: base saturation; m: Aluminum saturation.

With the results of the chemical analysis of the soil, according to the recommendations of the Soil Fertility Commission of the State of Minas Gerais (Alvarez & Ribeiro, 1999), *Brachiaria brizantha* is defined as having a high technological level, in which there was a need for liming and application of phosphorus and nitrogen, with no need for the application of potassium.

After filling the pots, limestone was applied to the surface and remained in incubation for 30 days to neutralize the soil acidity. During incubation, the soils were maintained at 80% of their field capacity, with replacement being carried out at one-day intervals.

The determination of field capacity was performed using all pots with dry soil and weighed, soaked with water and, after the water had completely drained, weighed again. This weight corresponds to the weight of the soil close to field capacity, and was used to replace water lost through evapotranspiration, with all these pots being weighed daily.

After the incubation period in the pots, *Brachiaria brizantha* cv Braúna was planted, using approximately 20 seeds per pot, at a depth of approximately 2 cm. Liquid and conventional fertilization was carried out 15 days after planting, and after each regrowth period, nitrogen fertilization was carried out in the remaining pots.

Nitrogen fertilization was divided into 3 applications, the first being carried out after sowing and the following after each cut, totaling 150 kg N ha<sup>-1</sup> corresponding to doses of 0.35 g pot<sup>-1</sup> of urea per application. Phosphorus application was carried out with an application of 110 kg of P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup> corresponding to 0.78 g pot<sup>-1</sup> of simple superphosphate.

For liquid fertilization, it was carried out according to the manufacturer's recommendation and soil analysis, where the following NPK formulas were used: 06:30:00 and 30:00:00. In the implementation, 1.07 mL of liquid NPK 06:30:00 + 0.4 mL of liquid NPK 30:00:00 was used, corresponding to 0.45 g of P<sub>2</sub>O<sub>5</sub> and 0.49 g of N, and in the maintenance, 0.47 mL of liquid NPK 06:30:00 + 0.52 mL of liquid NPK 30:00:00 was applied, which corresponds to 0.2 g of P<sub>2</sub>O<sub>5</sub> and 0.212 g of N.

After 30 days of seed germination, thinning was carried out, leaving 4 plants per pot, where the criterion used was the vigor and homogeneity of the plants. 20 days after thinning, a uniform cut was carried out at 10 cm above the soil and from the uniformity, the evaluations began.

At the end of each cutting period, four pots per treatment were dismantled, so that in each 21-day interval, 12 pots were dismantled, obtaining dismantled at 21, 42 and 63 days counted from the standardization cut. The roots were cleaned with running water and their volume and dry mass were evaluated. Before each dismantling, the soil was collected in each pot in the 0 to 20 cm layer, collecting it at four points in the pots, where they were combined to obtain the composite sample.

### **Plant analysis**

To assess dry matter, for each cut, 4 pots were dismantled with the aid of running water, removing the entire plants, which were later dissected into leaves, stems, roots and residue. The aerial part sample was considered above the recommended 10 cm cutting line, and the residue corresponded to the production below the cutting line, where the production data were presented in kg DM ha<sup>-1</sup>, considering the pot area of 0.07065 m<sup>2</sup>.

Immediately after cutting, the collected material was properly identified and taken to the Laboratory of Anatomy and Ecological Physiology of Plants and weighed for later determination of the dry matter production of the forage. To determine pre-drying, the identified material was weighed as a green sample and after pre-drying in a forced circulation oven at 55 °C for 72 hours, it was ground in a Willey knife mill with a 1 mm sieve. After grinding, the final dry matter was determined, following the methodology described by (Detmann et al., 2012).

To evaluate the roots, in addition to the dry matter, the volume was determined. For this purpose, a test tube with a known quantity of water was used, where the fresh root was introduced and, through the difference in observed volume, the root volume was obtained.

To evaluate the bromatological composition, the contents of dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ash were determined according to Detmann et al. (2012).

For the growth study, 2 tillers per pot were marked with colored tapes (green and red), where after the uniformity cut, the following characteristics were evaluated every three days, throughout the growth period: appearance of the leaf apex; stem length; number of leaves; leaf length and width.

Using the data obtained during the evaluations, the following characteristics were calculated:

- Leaf appearance rate (LAR, leaves/tiller/day): obtained by dividing the number of leaves that appeared on the marked tillers of each pot by the regrowth period;
- Leaf elongation rate (LER, cm/tiller/day): calculated by the difference between the final and initial leaf lengths, divided by the measurement interval;
- Leaf/leaflet width (LL, cm): average width of the leaf blades or of the three fully expanded leaflets;
- Final leaf length (FLL, cm): obtained by measuring the fully expanded leaves, from their ligule to the leaf apex.
- Stalk elongation rate (SER, mm/tiller/day): obtained by the difference between the final and initial length of the stalk of each tiller, measured from soil level to the height of the ligule of the youngest leaf, divided by the interval of measurements;
- Final stem length (CFC, cm);
- Total plant length (CTP, cm);

To quantify the leaf area, the leaves of the four plants, per pot, were scanned. All leaf blades were digitized using an HP G2710 photo scanner at 300 dpi resolution and subsequently analyzed using the ImageJ® program, with the leaf area (LA) being calculated and its value expressed in  $\text{cm}^2 \text{ pot}^{-1}$ . Based on the AF data, the leaf area index (LAI), specific leaf area (SLA) and leaf area ratio (LAR) were calculated according to the equation defined by Cairo et al. (2008).

At the end of the growth cycle, lasting 28 days, two fully expanded leaves were collected from each experimental unit and placed in envelopes made of aluminum foil and immediately stored on ice, which were taken to the laboratory for determination of the chlorophyll a, chlorophyll b and carotenoid contents, where 0.03 g of the fresh mass (FM) of the collected leaf was placed in a glass vial containing 5 mL of dimethyl sulfoxide and wrapped in aluminum foil for 72 hours. Then, a reading was performed on the spectrophotometer at 665, 649 and 480 nm of absorbance, to quantify the photosynthetic pigments with formulas defined by Wellburn (1994) and values adjusted to  $\text{mg g}^{-1} \text{ MF}$ .

### **Statistical analysis**

The data obtained were analyzed using the statistical program SAS - Free Statistical Software, SAS University Edition. Analysis of variance was performed, considering the limestone source, regrowth and the interaction between the limestone source and regrowth as sources of variation. The comparison between the effects of the limestone source and regrowth were performed using the *Tukey test*.  $\alpha = 0.05$  was adopted.

## RESULTS AND DISCUSSION

There was a numerical difference between conventional fertilization and liquid fertilization in the main soil components (phosphorus (P), calcium (Ca) and magnesium). Conventional fertilizer was the one that demonstrated the highest values for these components (Table 2).

**Table 2.** Effect of fertilization on soil fertility in the 0 to 20 cm layer after regrowth periods

FERTILIZING	mg dm <sup>-3</sup> P	Cmolc dm <sup>-3</sup> of soil						%
		K <sup>+</sup>	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Al <sup>3+</sup>	S.B.	T	V
21 days								
liquid	4.00	0.06	2.70	2.20	0.27	5.13	9.93	51.67
conventional	13.00	0.08	2.97	2.27	0.13	5.60	10.30	54.00
42 days								
liquid	7.00	0.05	2.17	2.00	0.40	4.40	10.93	40.33
conventional	15.00	0.04	2.87	2.00	0.30	5.10	10.73	47.33
63 days								
liquid	4.67	0.05	2.03	1.83	0.47	4.17	10.90	38.33
conventional	26.67	0.04	2.70	1.83	0.43	4.83	11.87	41.00

P = phosphorus; E = potassium; Com = Calcium; E = magnesium; S.B. = base saturation; T = cation exchange capacity; V = percentage of base saturation.

The analysis of phosphorus (P) levels in the soil revealed the superiority of conventional fertilization in all regrowths, with a progressive increase in each cycle. This result can be attributed to the low mobility of phosphorus in the soil, which limits its availability to plants when applied diluted in water, as in liquid fertilization. The leaching of this nutrient, resulting from liquid application, may have contributed to the lower efficiency of this fertilization method.

Considering that phosphorus is one of the most limiting mineral nutrients in tropical soils, replenishing this nutrient becomes essential to ensure pasture productivity. Phosphorus plays a crucial role in the storage, transport and utilization of energy during photosynthesis, in addition to participating in protein synthesis and enzymatic metabolism. Its influence on root growth and plant tillering makes it an essential element for the development of grasses, being considered the most important nutrient after nitrogen (Werner, 1984).

Analysis of potassium (K) levels in the soil revealed remarkable stability, with values close to 0.06 cmolc dm<sup>-3</sup> in all treatments, regardless of the type of fertilization or regrowth. This constancy can be attributed to the absence of potassium fertilization in the experiment, which limited the addition of potassium to the soil. Furthermore, the uniformity in potassium levels suggests that the consumption of this nutrient by plants was similar in all treatments, indicating that the different fertilization methods and regrowth cycles did not significantly influence the absorption of potassium by plants.

Analysis of the data in Table 1, which presents the results of the soil analysis performed prior to the start of the experiment, reveals a potassium (K) content of 0.41 cmolc dm<sup>-3</sup>, equivalent to 160.3 mg dm<sup>-3</sup> of K in the soil. This concentration of potassium in the soil is a crucial factor in determining the need for potassium fertilization.

According to Dias-Filho (2012), potassium fertilization in pasture formation is recommended only when the potassium content in the soil is less than 50 mg dm<sup>-3</sup>. The justification for this recommendation lies in the fact that high doses of potassium can lead to a reduction in the availability of magnesium (Mg) for plants. This antagonistic interaction between potassium and magnesium can compromise magnesium absorption by plants, resulting in nutritional deficiencies and negatively impacting pasture growth and development.

The analysis of calcium (Ca) levels in the soil revealed stability in the values throughout the three regrowths when conventional fertilization was applied. However, liquid fertilization resulted in a progressive decrease in Ca levels with each regrowth. It is important to highlight that all treatments received the same acidity correction with limestone, which suggests that the difference observed with liquid fertilization may be related to the dilution of the fertilizer in water, which may have favored calcium leaching.

Following a similar trend, the sum of bases (SB), cation exchange capacity (T) and base saturation (V) showed the highest values when conventional fertilization was applied. These results can be interpreted as a direct consequence of the greater availability of calcium in the soil, since calcium is a divalent cation that contributes significantly to the sum of bases and base saturation. The greater availability of calcium, therefore, resulted in an increase in the cation exchange capacity of the soil, reflecting in the greater capacity of the soil to retain cations and, consequently, in greater base saturation.

Data analysis did not reveal significant interaction ( $P > 0.05$ ) between the factors studied for dry matter production of the aerial part, residue and root, as well as for leaf/stem ratio and root volume (Table 3). This finding suggests that the effects of fertilization and regrowth on these variables were independent of each other.

However, shoot dry matter production was significantly influenced by both regrowth and fertilization. Conventional fertilization promoted the highest dry matter production results, demonstrating its superiority over liquid fertilization. Furthermore, the first two regrowths showed the highest dry matter production rates, indicating a reduction in productivity as the regrowth cycles progressed. This observation suggests that the plant's recovery capacity may decrease over time, or that soil nutrient availability may have been reduced during regrowth.

**Table 3.** Effect of regrowth and fertilization on the production of dry mass of aerial part (PMSPA), dry mass of residue (PMSRE), dry mass of root (PMSRA), leaf/stem ratio and root volume of *Brachiaria brizantha* cv. Braúna

Variables	Treatments					CV <sup>1</sup>	<i>p-value</i>		
	Regrowth			Fertilizing					
	1 <sup>a</sup>	2 <sup>a</sup>	3 <sup>a</sup>	Agreement	nal Liquid		da	R	A
PMSPA <sup>2</sup>	966.0	727.7	608.0 B	848.1 a	686.3 b	20.48	0.0008	0.021	0.617
PMSRE <sup>2</sup>	779.9 B	906.6B	1,603.6	1,101.3 a	1,092.0	17.36	< .0001	0.906	0.348
PMSRA <sup>2</sup>	1,032.9	873.4 A	1,159.1	1,029.5 a	1,014.1	33.47	0.2715	0.912	0.985
Reason eaf/stem	2.2 B	2.4 B	4.0 A	2.8 a	3.0 a	10.60	< .0001	0.124	0.714
Root volume <sup>3</sup>	170.0 B	195.0 B	268.8 A	207.5 a	215.0 a	19.16	0.0003	0.655	0.347

<sup>1</sup>Coefficient of variation in percentage. 2 kg ha<sup>-1</sup>. 3 mL. R = Regrowth; A = Fertilizing; RxA = interaction between factors. Averages followed by the same letter, in the same line, do not differ from each other by the test.

The practice of frequently cutting the aerial part of plants leads to a decrease in biomass production, a phenomenon attributed to the removal of photosynthetically active areas and consequent restriction of the accumulation of organic reserves. This condition predisposes the plant to less vigorous regrowth, as observed by Costa et al. (2014).

In the context of the present study, the reduction in the productivity of the aerial part of Braúna grass can be explained by the adopted cutting height of 10 cm. This practice differs from the recommendations for most *Brachiaria brizantha* cultivars, which recommend a residue height greater than 15 cm. Furthermore, the 21-day interval between cuts may have been insufficient for adequate forage recovery. For the Braúna cultivar, a rest period of 25 to 28 days is suggested to optimize regrowth and biomass production (Matsuda, 2016).

Data analysis revealed that conventional fertilization promoted a significant increase ( $p < 0.05$ ) in the production of dry mass of the aerial part of the grass. However, for the other variables evaluated, no significant differences ( $p > 0.05$ ) were observed between the two types of fertilization.

The higher dry mass production of the aerial part observed with conventional fertilization can be explained by the results of the soil analysis (Table 2). Liquid fertilization resulted in lower availability of essential nutrients, such as calcium, phosphorus and potassium, which may have limited plant growth and development. The lower availability of nutrients with liquid fertilization can be attributed to leaching, a process in which nutrients are removed from the soil by water, making them unavailable for absorption by plants.

The production of dry mass of the residue was not influenced by the type of fertilization ( $p > 0.05$ ), but was affected by regrowth ( $p < 0.05$ ), which demonstrated a greater result in the 3<sup>rd</sup> regrowth. This result is the opposite of the production of dry mass of the aerial part, making it clear that there was less development of leaves and stems and an increase in biomass at the base of the plant. This can be justified by the previously recommended cutting height of only 10 cm, which allowed new leaves to emerge from the basal buds, causing a greater accumulation of residue.

The type of fertilization and regrowth did not influence the production of root dry mass ( $p > 0.05$ ), which demonstrated stability in growth for both regrowth and the type of fertilization. Temporary paralysis or reduction in the growth rate of the root system may occur due to the mobilization of root reserves for recovery of the aerial part (Cecato et al., 2004). Therefore, the reduction in root growth after defoliation can be considered as an adaptation mechanism, which allows for a faster reestablishment of the leaf area and the eventual restoration of the balance between root and shoot growth (Richards, 1984).

The leaf/stem ratio was influenced by regrowth ( $p < 0.05$ ), with a higher ratio in the 3<sup>rd</sup> regrowth. The higher the ratio, the greater the proportion of leaf in relation to the stem, thus implying better quality forage. This effect can be explained by the growth dynamics of the aerial part, which resulted in a lower stem elongation (LER) and the leaf appearance rate was maintained (RApF), which leads to a higher ratio. The higher this leaf/stem ratio, the better, since the leaf is one of the morphological components with the best nutritional value (Fonseca & Santos, 2009).



Root volume was affected only by regrowth ( $p < 0.05$ ), showing greater volume in the 3<sup>rd</sup> regrowth. Intense cutting of the aerial part reduces root growth, but to compensate for this effect, the plants modified the morphology of the root system, increasing the area available for nutrient absorption through the formation of root hairs (Thornton & Millard, 1996). This explains the greater volume with the increase in cuts without changing total root production.

Data analysis revealed the absence of significant interaction ( $p > 0.05$ ) between the factors studied for all variables related to plant morphology (Table 4). This result suggests that the effects of regrowth and fertilization on morphological characteristics were independent of each other.

**Table 4.** Effect of regrowth and fertilization on the leaf appearance rate (RApF), final leaf width (FLW), final leaf length (LFL), leaf elongation rate (LER), final stem length (FSL), stem elongation rate (SER) and total plant length (TPL) of *Brachiaria brizantha* cv. Braúna

Variables	Treatments					CV <sup>1</sup>	<i>p-value</i>		
	Regrowth			Fertilizing					
	1 <sup>a</sup>	2 <sup>a</sup>	3 <sup>a</sup>	Conventional	Liquid		R	A	RxA
RApF	0.098	0.098	0.098	0.099	0.105	15.385	1.0000	0.0571	0.2188
FLW	1.34 A	1.21 AB	0.976 B	1.180	1.172	12.030	0.0002	0.8822	0.7418
LFL	30.71A	23.02 B	19.13 C	25.11 a	23.47 b	6.410	<0.0001	0.0188	0.9922
LER	2.99 A	2.55 AB	1.93 B	2.65	2.33	21.790	0.0038	0.1635	0.0792
FSL	18.56 A	20.09 A	13.52 B	17.33	17.45	16.990	0.0008	0.9267	0.8640
SER	0.41A B	0.52 A	0.26 B	0.39	0.404	29.900	0.0010	0.7683	0.7394
TPL	47.25 A	42.88 A	34.94 B	44.04 a	39.33 b	10.620	0.0001	0.0179	0.1070

<sup>1</sup>Coefficient of variation in percentage. R = Regrowth; A = Fertilizing; R×A = interaction between factors. Means followed by the same letter, in the same line, do not differ from each other by the *Tukey test* ( $p > 0.05$ ).

However, regrowth significantly influenced ( $p < 0.05$ ) all morphogenetic variables evaluated, with the exception of the rate of leaf appearance, which remained constant throughout the cuts. This observation indicates that the dynamics of plant growth and development were affected by the regrowth cycle, with variations in height, length and number of tillers.

Fertilization, in turn, significantly influenced ( $p < 0.05$ ) the final leaf length (LFL) and total plant length (TPL). This result suggests that the availability of nutrients provided by fertilization affected the growth and development of the aerial part of the plant, resulting in longer leaves and taller plants.

The rate of leaf appearance was not influenced by the type of fertilization and regrowth ( $p > 0.05$ ), with conventional fertilization showing the best results. Morphogenic variables are affected by the availability of growth resources such as water, light, nitrogen and temperature (Diafante et al., 2008). After cutting, plants seek to rebuild their leaf area in order to maximize the interception of incident light. At this stage of canopy growth, there is no competition for light and, therefore, the plant prioritizes the production of leaf tissue, which justifies the constancy in the leaf appearance rate (RApF) in all regrowths.

For final leaf width, final leaf length, leaf elongation rate, final stem length, stem elongation rate and total plant length, there was an effect of regrowth for all these characteristics ( $p < 0.05$ ).

According to Moore & Moser (1995), there is a direct relationship between the size of the pseudostem, the speed at which leaves appear and the length of these leaves. This occurs because young leaves develop inside the cartridge formed by the sheaths of older leaves. Thus, the greater the length of this cartridge (greater height of the pseudostem), the longer the young leaves remain stretching inside it. As a result, if the cartridge is small, the leaves appear faster and are smaller leaves. If the cartridge is long, the leaves take longer to elongate, take longer to appear and their final size is larger. Thus, after cutting, the leaves appear quickly (high appearance rate), but each leaf is small, since the pseudostem is short, which justifies the results found in this study for the LFL and LAR variables, which despite having a constant leaf appearance rate, the elongation rate and final leaf length decreased with each regrowth.

The interaction between regrowth and fertilization was significant ( $p < 0.05$ ) for leaf area, specific leaf area, leaf area index and leaf area ratio of *Brachiaria brizantha* cv. Braúna grass (Table 5).

**Table 5.** Effect of regrowth and fertilization on leaf area (LA), specific leaf area (SLA), leaf area index (LAI) and leaf area ratio (LAR) of *Brachiaria brizantha* cv. Braúna

Regrowth	Treatments		Average	CV <sup>1</sup>	<i>P-value</i>		
	Liquid	Conventional			R	A	RXA
LA							
21 days	2,144.63 Bb	3,438.70 Aa	2,791.67	7.79	< .0001	0.0003	< .0001
42 days	2,613.53 Aa	2,356.22 Ba	2,484.88				
63 days	1,918.72 Ba	1,911.09 Ca	1,914.91				
Average	2,225.63	2,568.67					
SLA							
21 days	39.49 Bb	47.62 Aa	43,555	16.15	0.2636	0.2522	0.0392
42 days	54.33 Aa	41.84 Bb	48,085				
63 days	53.14 Aa	46.46 Ba	49.8				
Average	48.99	45.31					
LAI							
21 days	3.04 Bb	4.87 Aa	3.95	7.79	< .0001	0.0003	< .0001
42 days	3.70 Aa	3.34 Ba	3.52				
63 days	2.72 Ba	2.71 Ca	2.71				
Average	3.15	3.64					
LAR							
21 days	7.89 Ab	10.62 Aa	9.25	14.44	< .0001	0.2874	0.0016
42 days	8.17 Aa	6.54 Ba	7.36				
63 days	4.21 Ba	4.47 Ba	4.34				
Average	6.76	7.21					

<sup>1</sup>Coefficient of variation in percentage. 2 kg ha<sup>-1</sup>. 3 mL. R = Regrowth; A = Fertilizing; R×A = interaction between factors. Means followed by the same capital letter in the column and lowercase in the row do not differ from each other by the *Tukey test* ( $p > 0.05$ ).

Plants that received conventional fertilization showed greater leaf area and leaf area index in the first regrowth, while those that received liquid fertilizer showed a higher value in the second regrowth. When comparing this variable to the type of fertilization received throughout the regrowth, it can be seen that conventional fertilization was superior only in the first regrowth and in the other regrowths there was no difference

between the type of fertilization. In addition, it can be observed that there was a reduction in leaf area throughout the regrowth, regardless of the type of fertilization.

For the specific leaf area, when liquid fertilizer was applied, the first regrowth was lower, while for conventional fertilizer, the first and third regrowths were higher. This may have occurred due to competition for light, stress caused by cutting and the availability of nutrients, which was lower for liquid fertilization, since species tend to change their anatomical characteristics in order to optimize light capture (Furquim et al., 2013).

The LAI, in general, decreased with each regrowth for both types of fertilizer. These results were mainly due to the high cutting intensity. Grasses cut under high intensity present remarkable morphogenic responses such as a higher leaf appearance rate, which in turn may not reflect an increase in leaf area index and forage accumulation, as these plants generally present a lower final leaf length, characterizing the capacity for morphophysiological adaptation represented as phenotypic plasticity of the canopy (Barbosa et al., 2011).

Regrowth had an effect on LAR, in both fertilizations the leaf area ratio decreased throughout the regrowth, due to the lower LA and higher total MS during the regrowth, since LAR is determined by the formula  $LAR = AF_{total}/MS_{total}$ . When considering the fertilizations, there was a difference between them only in the 1st regrowth, due to the difference in AF in this period, with conventional fertilizer being superior to liquid fertilizer, making it evident that regrowth had a greater influence on this variable than the type of nutrient availability for both fertilizations.

The interaction was not significant ( $p > 0.05$ ) for the SPAD index, chlorophyll a, chlorophyll b and total (Table 6). There was an effect of regrowth for the SPAD index, chlorophyll b content and total chlorophyll ( $p < 0.05$ ). Fertilization influenced only the SPAD index ( $p < 0.05$ ).

**Table 6.** Effect of regrowth and fertilization on SPAD index, chlorophyll a, chlorophyll b, and carotenoid content of *Brachiaria brizantha* cv. Braúna

Variables	Treatments					CV <sup>1</sup>	<i>p-value</i>		
	Regrowth			Fertilizing					
	1 <sup>a</sup>	2 <sup>a</sup>	3 <sup>a</sup>	Conventional	Liquid		R	A	RxA
SPAD	32.529b	34.963a	36.067a	35.53A	33.51B	6.490	0.0163	0.0413	0.8718
Chlorophyll a	1.02	1.08	1.18	1.11	1.08	13.710	0.1300	0.6451	0.0629
Chlorophyll b	0.452 b	0.670 a	0.702 a	0.57	0.64	14.600	<0.0001	0.0594	0.9704
Total	1.475 b	1.752 ab	1.884 a	1.68	1.72	13.86	0.0087	0.5975	0.3289

<sup>1</sup> Coefficient of variation in percentage. R = Regrowth; A = Fertilizing; RxA = interaction between factors. Means followed by the same letter, in the same line, do not differ from each other by the *Tukey test* ( $p > 0.05$ ).

For the SPAD index, there was an effect of regrowth and fertilization. An increase in the value from the 1<sup>st</sup> to the 2<sup>nd</sup> and 3<sup>rd</sup> regrowth was observed for this variable, which can be justified by the maintenance nitrogen fertilization applied at each cut. When comparing the type of fertilization, the one that provided the highest index was conventional fertilization, which can be explained by the greater nitrogen assimilation that the fertilization provided, and this can be proven by the bromatological analysis (Table 7), which demonstrated a higher protein value in the grass that received conventional fertilization.

The SPAD index is a unit of measurement expressed by the chlorophyll meter, which corresponds to the chlorophyll content present in the leaf (Rocha et al., 2005). This measurement is related to the N content (Rocha et al., 2005; Barbieri Junior et al., 2012) and crude protein in the leaf (Maranhão et al., 2009), enabling the early diagnosis of a possible deficiency. This explains the increase in the SPAD index in the second and third regrowth, demonstrating that maintenance fertilization with conventional fertilizer was more efficient than with liquid fertilizer.

Chlorophyll type a was not affected by fertilization or regrowth ( $p > 0.05$ ), maintaining an average value of  $1.10 \text{ mg g}^{-1}$ . Chlorophyll *a* is the pigment used to carry out photochemistry (the first stage of the photosynthetic process), while the other pigments assist in the absorption of light and the transfer of radiant energy to the reaction centers, thus being called accessory pigments (Streit et al 2005).

For chlorophyll *b*, there was an effect of regrowth for this variable ( $p < 0.05$ ), where an increase in values can be observed with each cut made to the plant. This increase in chlorophyll *b* is used as a compensatory mechanism to assist chlorophyll *a* in the photosynthetic processes.

There was an interaction between regrowth and type of fertilization on the chlorophyll *a:b* and carotenoid ratio ( $p < 0.05$ ). For conventional fertilization, the 1st regrowth showed the highest value of this ratio. For liquid fertilization, there was a difference in the 2nd regrowth, which was smaller than the 1st and 3<sup>rd</sup> regrowths, which was the lowest ratio found (Table 7). This difference found in the second regrowth for liquid fertilization is due to the increase in chlorophyll type *b* in this period.

**Table 7.** Effect of regrowth and fertilization on the chlorophyll *a:b* ratio and carotenoids of *Brachiaria brizantha* cv. Braúna

Regrowth	Fertilizing		Average	CV <sup>1</sup>	<i>p</i> -valor		
	Conventional	Liquid			R	A	RxA
Chlorophyll a:b ratio							
1 <sup>a</sup> regrowth	2.80Aa	1.82Ab	2.31	3.67	< 0.0001	< 0.0001	< 0.0001
2 <sup>a</sup> regrowth	1.72Ba	1.53Bb	1.62				
3 <sup>a</sup> regrowth	1.67Ba	1.70Aa	1.69				
AVERAGE	2.06	1.68					
Carotenoids							
1 <sup>a</sup> regrowth	0.31 Ab	0.40 Ba	0.355	8.86	0.5011	< 0.0001	0.0017
2 <sup>a</sup> regrowth	0.27 Ab	0.43Aba	0.350				
3 <sup>a</sup> regrowth	0.25 Ab	0.48 Aa	0.365				
AVERAGE	0.28	0.44					

<sup>1</sup>Coefficient of variation in percentage. R = Regrowth; A = Fertilizing; RxA = interaction between factors. Means followed by the same letter, capitalized in the column and lowercase in the row, do not differ from each other by the *Tukey test*.

Chlorophyll *a* and *b* are found in nature in a ratio of 3:1 (Streit et al., 2005). The chlorophyll *a:b* ratio was lower than that described in the literature, which can be justified by the fact that the plants suffered severe and successive cuts in a short period of time, which led to an increase in the chlorophyll *b* content as a compensation mechanism and therefore decreasing this ratio.

For carotenoids, there was no effect on regrowth when conventional fertilizer was applied, maintaining an average of 0.28 mg g<sup>-1</sup> of fresh matter. For liquid fertilizer, the carotenoid levels increased with each regrowth, that is, for plants that received liquid fertilizer, there was an increase in accessory pigments (chlorophyll b and carotenoids) to help in the reestablishment of these plants.

There was an effect of fertilization on carotenoid levels ( $p < 0.05$ ), regardless of regrowth, this pigment was more present when liquid fertilizer was applied. Carotenoids play a fundamental role in protecting thylakoid membranes, because in addition to being light collectors, they act as photoprotective pigments, preserving chlorophylls from the oxidative destruction of O<sub>2</sub>, when there is excess energy (Silva et al., 2001).

The interaction was not significant ( $p > 0.05$ ) for the bromatological variables dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF) and ash (Table 8).

**Table 8.** Effect of regrowth and fertilization on the chemical composition of *Brachiaria brizantha* cv. Braúna

Variables	Treatments					CV <sup>1</sup>	<i>p-value</i>		
	Regrowth			Fertilizing					
	1 <sup>a</sup>	2 <sup>a</sup>	3 <sup>a</sup>	Conventional	Liquid		R	A	RxA
DM (%)	20.974 b	20.291b	23.239	21.399	21.603	3.070	< 0.0001	0.4605	0.4848
CP (%)	9.759	10.142	9.931	10.465 A	9.422 B	8.870	0.6898	0.0096	0.0968
NDF (%)	68.59a	71.46ab	74.99 b	70.928	72.430	4.370	0.0027	0.2561	0.7922
ADF (%)	34.2 a	36.681ab	37.174	35.798	36.238	6.710	0.04	0.6610	0.9768
Ashes (%)	6.988 a	6.383 ab	6.219 b	6.526	6.534	6.290	0.0037	0.9613	0.1898

DM: dry matter; CP: crude protein; NDF: neutral detergent fiber; ADF: acid detergent fiber; <sup>1</sup>coefficient of variation; R: regrowth; A: fertilization. Means followed by the same letter, in the line, do not differ from each other by the *Tukey test* ( $p > 0.05$ ).

The dry matter content was not influenced by the type of fertilization ( $p > 0.05$ ), but was affected by regrowth ( $p < 0.05$ ), showing a higher DM value in the third regrowth. This result can be explained by the successive cuts that the plant suffered, causing acceleration in the photosynthetic processes.

For crude protein, there was no effect of regrowth ( $p > 0.05$ ), but it was influenced by fertilization ( $p < 0.05$ ), with conventional fertilization increasing PB by 1.04% compared to liquid fertilization. According to Oliveira et al. (2010), the nutritional value of forage (including crude protein) can be altered by temperature, water, soil fertility, among others. Therefore, the increase in crude protein with the use of conventional fertilizer was due to the greater availability of nutrients in the soil for this treatment, as shown in Table 2.

In the variables NDF, ADF and ash, there was an effect of regrowth ( $p < 0.05$ ). For NDF and ADF, there was an increase in these levels from the first to the third regrowth, obtaining in the third regrowth NDF and ADF levels of 74.99 and 37.17%, respectively. Results similar to those found by Santos & Santos (2020), working with the same cultivar and cutting interval, found values of 72.88 and 35.84% for NDF and ADF. These results may be a consequence of the stress caused by successive cuts of 21 days and a height of only 10 cm from the ground.

The NDF content is related to the consumption and quality of the dry mass of a forage; as the NDF increases, the amount of dry mass ingested decreases. According to Van Soest (1965), values above 55–60% of NDF directly influence the consumption of the forage. On the other hand, the ADF indicates a greater proportion of components resistant to digestion, therefore, directly affecting the digestibility of a forage.

For ash, the effect was the opposite; with the increase in regrowth there was a decrease in the amount of ash, explained by the increase in NDF and ADF, since these components of organic matter increase, generating a decrease in the mineral fraction and moisture of the forage.

## CONCLUSIONS

Liquid fertilizer does not provide improvements in soil fertility, dry matter production and crude protein of Braúna grass, and is not recommended as a substitute for conventional fertilizer, negatively influencing regrowth and, consequently, the productive and morphophysiological characteristics of *Brachiaria brizantha* cv. Braúna grass.

For this grass, successive cuts every 21 days and 10 cm from the soil are not recommended, as they interfere with the reestablishment of the grass. Further studies are required with cutting intervals greater than 21 days and residue heights above 10 cm for this cultivar.

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