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Effect of Organic Fertilization on the Production and Nutritional Quality of *Brachiaria Decumbens* in the Ecuadorian Amazon

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ABSTRACT

Introduction: Soil fertility is key to agricultural productivity. The Ecuadorian Amazon, one of the most biodiverse regions on the planet, is home to tropical ecosystems of great ecological importance. Agricultural activity, centred mainly on livestock, has grown in recent decades due to the high demand for meat and derived products.

Objectives: To evaluate the impact of different levels of biofertilizer biol based on previous research and its effect on the growth and nutritional quality of the grass species *Brachiaria decumbens*.

Methods: The experiment was conducted at the Centro Experimental de Investigación y Producción Amazónica (CEIPA). Universidad Estatal Amazónica (UEA), using a randomized block design with three treatments (T1, T2, T3) and a control without fertilizer. The different doses of biol were applied every 15 days and biomass sampling was carried out at 15, 30 and 45 days. The parameters evaluated included morphological variables such as plant height, number of stems and leaves, stem diameter and leaf dimensions, as well as bromatological analyses measuring crude protein, fat, fiber, ash and dry matter.

Results: The results showed that the biol dose (T3) was the most effective in promoting plant growth, with an average height of 78.77 cm, with 82 stems per plant, five leaves per stem, and a leaf size of 33 cm. In addition, T3 presented significantly higher levels of protein (12.27 %), fat (5.19 %), fiber (28.20 %), ash (7.87 %) and dry matter (33.19 %).

Conclusions: These results indicate that the use of biol not only increases pasture productivity, but also improves its nutritive value, contributing to a more balanced diet for cattle in regions with poor and eroded soil.

Keywords: Bromatological analysis, Morphological parameters, Forage, Biol.

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INTRODUCTION

The Ecuadorian Amazon, one of the most biodiverse regions of the planet, harbors tropical ecosystems of great ecological importance. However, it faces serious challenges in the sustainable use of its natural resources, especially in agriculture (Viteri-Salazar and Toledo, 2020). Agricultural activity, mainly focused on livestock, has grown in recent decades due to the high demand for meat and derived products (Soto-Cabrera et al., 2020).

In this context, forage production is key to sustain livestock feeding. One of the most widely used grass is *Brachiaria decumbens* (dallis grass), valued for its tolerance to tropical climate and poor soils (Muniandy et al., 2020). However, its productivity is limited by low soil quality, nutrient deficiencies and overexploitation (Conrado et al., 2024). This situation calls for sustainable alternatives, as the excessive use of chemical fertilizers has deteriorated soil and affected the environment.

In response to this, organic fertilizers have gained importance. Among them, biol, a liquid resulting from anaerobic fermentation of organic wastes has been shown to improve soil fertility, promote plant growth and favor microbial activity (Condo and Ulloa, 2019; Vera- Cedeño et al., 2024). However, its application in Amazonian forages still requires further research, due to the scarcity of specific studies in this region.

Cattle ranching in the Amazon faces difficulties in maintaining productive pastures in a sustainable manner. The indiscriminate use of synthetic inputs has led to soil degradation and contamination of water sources (Tigrero-Zapata et al., 2022). Therefore, there has been growing interest in more environmentally friendly agricultural practices, such as the use of natural fertilizers. Amazonian soils, generally acidic, poor in nutrients and prone to erosion, hinder plant growth (González et al., 2019). Organic fertilization could be key to improving forage quality, especially in sensitive species such as *Brachiaria decumbens*.

In addition, many farmers lack access to adequate inputs and technical training, which limits the implementation of sustainable practices (González-Marcillo et al., 2023). This situation affects pasture productivity, negatively impacting livestock profitability and ecosystem conservation.

This study is framed within the need to identify sustainable alternatives for forage production in the Amazon. The application of biol is proposed as a viable strategy to improve the morphological and bromatological characteristics of *Brachiaria decumbens*, contributing to a more profitable and environmentally responsible livestock production. The objective of this study was to evaluate the effect of different levels of biol on the morphological and bromatological variables of *Brachiaria decumbens* grass in the Ecuadorian Amazon.

OBJECTIVES

To evaluate the impact of different levels of biofertilizer biol based on previous research and its effect on the growth and nutritional quality of the grass species *Brachiaria decumbens*.

METHODS

The experiment was conducted at the Centro Experimental de Investigación y Producción Amazónica (CEIPA) of the Universidad Estatal Amazónica, located in zone 18 of Puyo, Pastaza, Ecuador (UTM coordinates: 178879 m E, 9863155 m S). The region has a tropical climate, with average annual precipitation of approximately 4000 mm, relative humidity of 80 %, temperatures between 15 °C and 25 °C, gently undulating topography and an altitude located between 590 and 957 m asl (INAMHI, 2015).

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Table 1. Soil types of the Centro Experimental de Investigación y Producción Amazónica (CEIPA).

Provinces	Order	Suborder	Texture	Characteristics
Napo	Entisol	TIPYC UDIFLUVENTS	Loamy	Recent soil, no have formed horizons, low organic matter content.
Pastaza	Inceptisol	TROPEPT + ANDEPT	Clay loam	Shallow to moderately deep, sloping to very steep topography. Humid tropical climate soils

Source: (INIAP, 2020)

Table 2. Main climatic factors in the Centro Experimental de Investigación y Producción Amazónica (CEIPA).

Provinces	Precipitation	Temperature	Humidity relative	Wind speed	Wind chill
Napo	3600 mm	21.6°C	82%	5.6 km/h	21°C
Pastaza	4604 mm	22°C	95 %	5 km/h	22°C

Source: (INAMHI, 2015)

Experiment design

A completely randomized block design was used (due to the topographic characteristics of the experimental area and moisture contents), with four treatments and a control reference, each with three replications, totaling 12 experimental units. Each unit consisted of an area of 25 square meters, delimited with a separation of 1 meter between them to prevent interference between treatments.

Treatments

The treatments applied were:

- T0: Control, water only (5 L of water).
- T1: 0.5 L of biol diluted in 4.5 L of water.
- T2: 1.0 L of biol diluted in 4.0 L of water.
- T3: 1.5 L of biol diluted in 3.5 L of water.

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Compositional characteristics of the biol

pH: The pH of the biol was in the range of 6 to 8, making it slightly acidic to neutral. A proper pH is important for nutrient availability and microbial activity in the soil.

Electrical conductivity: The electrical conductivity of the biol ranged from 1 to 3 mS/cm. This measure is related to the concentration of soluble salts in the solution and helps to evaluate its fertilizer potential.

Cation exchange capacity (CEC): The biol had a moderate cation exchange capacity, positive for improving nutrient retention in the soil, favoring plant growth.

The volume of biol used in each treatment was prepared by mixing the specific amount in a 20-liter drum, respecting the dilution ratio and in accordance with previous research carried out at the Research Center.

It is worth mentioning that chemical fertilization was not used as a positive control because the research is focused on evaluating organic or sustainable agriculture methods, the use of chemical fertilizers is not aligned with the principles of the research.

Biol preparation and application

The biol was produced by anaerobic fermentation of organic wastes (agricultural and organic wastes from the research center) under controlled conditions, following standardized protocols to obtain a microbiologically active liquid. Before application, the biol was mixed with water in the proportion indicated for each treatment. The solution was applied in the morning, using manual sprinklers, ensuring uniform coverage over the surface of the pasture.

Sample collection

The grass was already established 3 years ago, so a uniformization cutting of *Brachiaria decumbens* was performed at the beginning of the study to standardize growth conditions. Subsequently, biomass samples of 500 g per experimental unit, selected from 10 randomly chosen plants, were collected every 15 days. This procedure was repeated throughout the trial, ensuring the representativeness of the sample at each stage of plant development.

Application of treatments:

Starting from establishment, every 15 days the treatments were applied at the respective doses, spraying uniformly over the entire surface of the assigned area.

Applications were always made in the morning to avoid losses due to evaporation.

Sample collection and variables evaluated Biomass was cut from the entire surface of each experimental unit at the beginning (day 0) and every 15 days (days 15, 30 and 45) to obtain samples of 500 g, randomly selected from 10 plants per unit. The sample comprised the representative plants of the area, cut at ground level.

Morphological variables evaluated included:

Plant height (cm): measured from ground level to the apex of the plant, using a tape measure.

Number of leaves per plant (leaves/plant): Counting foliar leaves on 10 randomly selected plants in each unit.

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Leaf length (cm): Measured from base to apex on five representative leaves per plant with a ruler.

Leaf width (cm): Measured in the middle part of the leaf on the same five leaves, with a caliper.

Stem diameter (cm): Taken in the middle third of the main stem with a caliper.

Samples for bromatological analysis were processed to determine the percentages (%) of:

Crude protein: By Kjeldahl method (AOAC, 2023).

Crude fat: Soxhlet extraction.

Crude fiber: By standard gravimetric method.

Ash: By muffle incineration at 550 °C.

Dry matter: By drying in an oven at 105 °C until constant weight.

Statistical analysis

The data collected were processed with Origin Pro-version 2022 software, applying descriptive statistical analysis and mean comparison tests (ANOVA), considering a significance level of $p < 0.05$ to determine differences between treatments.

Recommendations for future research It is suggested to keep accurate records of the quality of the biol, adjust the doses according to the stage of grass development and soil conditions, as well as to extend the study period to evaluate long-term effects. In addition, the incorporation of microbiological analyses of the biol in each application could provide additional information on the relationship between microbiota and plant growth.

RESULTS AND DISCUSSION

Morphological variables

The effects of the biol on the morphological variables of *Brachiaria decumbens* are presented in Figures 1 and 2. In general, a progressive increase in height, number of stems, stem diameter, number of leaves, and leaf dimensions was observed between 15 and 45 days, evidencing the positive effect of the biol on the structural development of the forage.

In plant height (Figures 1A and 1B), the T3 treatment showed sustained growth, going from 32.07± 0.76 cm at 15 days to 78.77± 0.50 cm at 45 days, far exceeding the control T0 (48.73± 0.35 cm). Similarly, the number of stems per plant (Figures 1C and 1D) increased at T3 from 28 ± 3 to 82 ± 4, reflecting greater tillering. As for stem diameter (Figures 1E and 1F), T3 maintained continuous growth, indicating an improvement in plant robustness.

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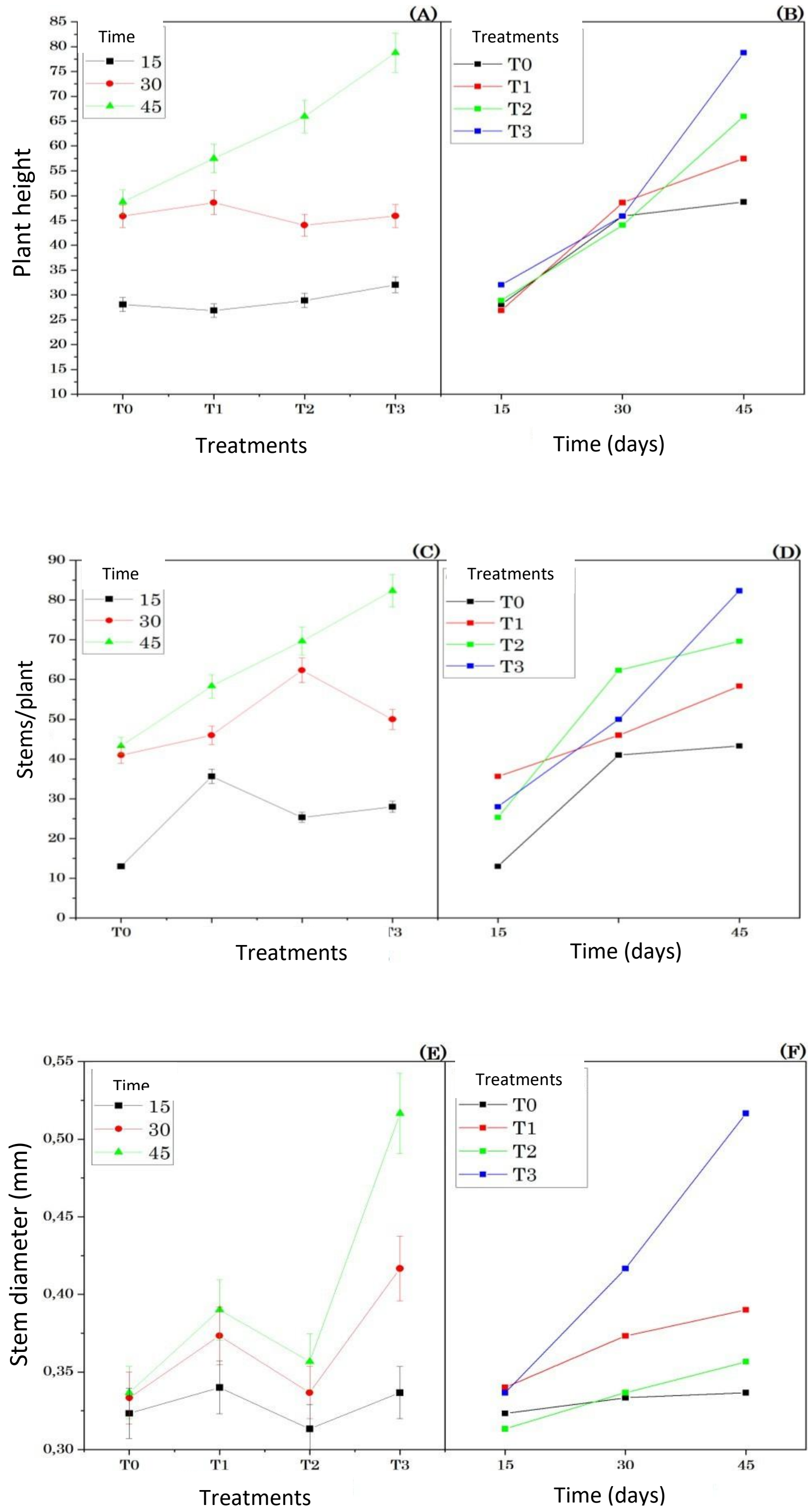

Figure 1. Effect of treatments and time on height, number of stems and stem diameter in B. decumbens.

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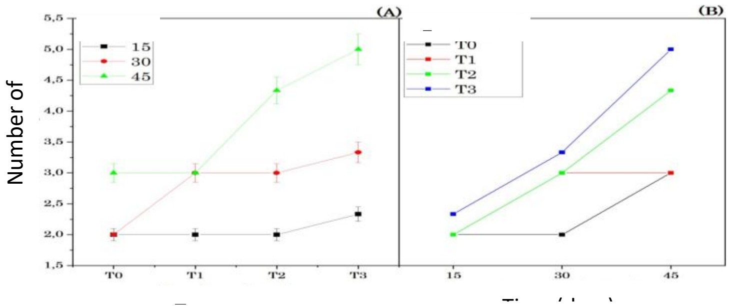
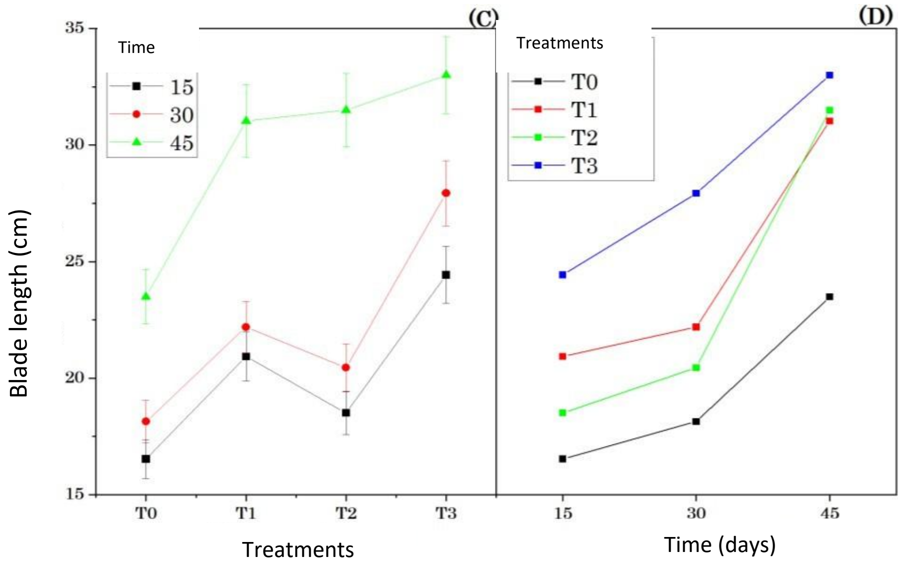
These results are in line with Techio-Pereira et al. (2022) and Edouard-Rambaut et al. (2022), who demonstrated that organic biofertilizers can improve the physiological efficiency of tropical grasses, thanks to an increase in the availability of essential nutrients such as nitrogen (N), phosphorus (P) and potassium (K), as well as an improvement in soil microbiological activity. Gil-Ramírez et al. (2023) also point out that biol contains hormonal compounds and beneficial microorganisms, such as nitrogen- fixing bacteria and mycorrhizal fungi, which stimulate plant growth and cell division, favoring tissue elongation and densification. In addition, biol, as a biofertilizer containing beneficial microorganisms, can improve the availability of nutrients in the soil by facilitating their solubilization and assimilation by the roots. The joint action of these microorganisms and organic nutrients favors root activity, which is reflected in the increase in height, number of stems and stem diameter in treated plants (T3). The increased root activity improves water and nutrient absorption, promoting vigorous growth. Regarding leaves, the number per stem increased significantly in T3, from 2 ± 0 to 5 ± 0 between 15 and 45 days (Figure 2A and 2B), surpassing the control that barely reached 2± 0 leaves per stem. Leaf length was also higher in T3, going from 24.43 ± 0.43 cm to 33 ± 0.62 cm (Figure 2C and 2D), while in T0 it was recorded only up to 23.5 ± 0.52 cm. In the case of leaf width, the T3 treatment showed an increase from 1.44 ± 0.03 cm to 1.63 ± 0.03 cm (Figure 2E and 2F), compared to the lower values of the control (1.36 ± 0.02 cm to 1.42 ± 0.03

cm).





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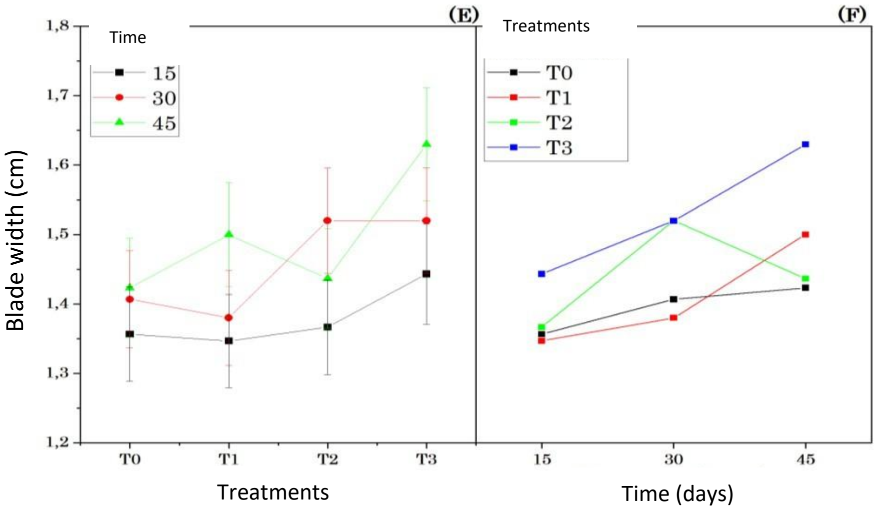

Figure 2. Effect of treatments and time on number of leaves per stem, leaf length and leaf width in B. decumbens.

The behavior observed in T3 could be attributed to a better carbon: nitrogen (C:N) ratio, the effective use of biol in forage production in the Ecuadorian Amazon, it is recommended that the C:N ratio of the biosubstrate or treated soil be in the approximate range of 20:1 to 25:1, adjusting according to the specific soil conditions, the composition of the biol and the crop cycle, to maximize the efficiency in the utilization of nutrients and achieve a healthy and productive growth of the pasture, a key aspect for the efficiency of biofertilizers according to Klumpp (2021) and Bi et al. (2023) who state that a balanced C:N favors the gradual release of nutrients and their utilization by the plant, minimizing leaching losses. In addition, Ji et al. (2024) and Ren et al. (2024) emphasize that the interaction between nitrogen-fixing bacteria and mycorrhizal fungi can improve soil structure and increase nutrient uptake, essential factors in tropical soils such as those of the Ecuadorian Amazon.

The results indicate that higher levels of biol (T3) generate a more pronounced effect on the variables evaluated, probably because the higher dose provides a higher concentration of beneficial microorganisms and organic nutrients, promoting a more effective interaction in the soil and in the plant. However, the trend of the results suggests that there is a dose-response relationship up to a certain point, after which there may be no additional benefits or even adverse effects.

Bromatological variables

Table 3 presents the results of the bromatological analyses at 15, 30 and 45 days. At 45 days, treatment T3 showed the highest protein (12.27 %), fat (5.19 %), fiber (28.20 %), ash (7.87 %) and dry matter (33.19 %) contents, compared to the progressively lower values observed in T0, T1 and T2. This trend was maintained at 30 days, where T3 continued to lead with 11.40 % protein, 4.29 % fat, 30.91 % fiber, 8.80 % ash and 30.79

% dry matter. At 15 days, although a slight decrease in protein and fat was evident in all treatments, T3 continued to lead with 11.40 and fat in all treatments, T3 retained its superiority, registering 10.12 % protein, 4.38 % fat, 32.41 % fiber, 9.11 % ash and 27.17 % dry matter.

Table 3. Chemical composition of *Brachiaria decumbens* at different cutting ages.

Age (days)	Treatment	% Crude protein	% Crude fat	% Crude fiber	% Total ashes	% Dry matter
15	T_0	10,41	3,80	35,76	9,52	25,13
15	T_1	10,10	4,22	32,73	9,80	27,58
15	T_2	10,62	4,31	32,69	9,92	27,88
15	T_3	10,12	4,38	32,41	9,11	27,17
30	T_0	11,22	4,00	30,35	8,64	27,37
30	T_1	11,15	4,04	30,60	8,93	30,94
30	T_2	11,25	4,27	30,93	8,10	30,22
30	T_3	11,40	4,29	30,91	8,80	30,79
45	T_0	12,15	4,12	28,17	7,92	27,26
45	T_1	12,25	5,72	28,59	7,70	33,37
45	T_2	12,09	5,15	28,69	7,10	33,85
45	T_3	12,27	5,19	28,20	7,87	33,19

Source: Authors

These findings indicate that the T3 treatment optimizes nutrient assimilation and forage structural development, confirming its superior efficacy compared to the other treatments. In particular, the protein content of 12.27 % and fat level of 5.19 % obtained in T3 at 45 days exceed the values reported by Reyes-Pérez et al. (2022) and Avellaneda-Cevallos et al. (2008), who found protein contents of 11.82 % and fat of 3.48 %, respectively. Likewise, the concentration of total fiber and ash at T3 was higher than that reported by Zemene et al. (2020), showing a substantial improvement in the bromatological quality of the forage after the appropriate application of biol. However, this same author reported a dry matter value (94.92 %) was considerably higher than that obtained in this study, a difference possibly attributable to factors such as the phenological stage of the forage, the edaphoclimatic conditions or the drying methods employed. The discrepancies observed with respect to the literature could also be due to the specific chemical composition of the biol used, as well as to the particular soil conditions, climate and agronomic management applied in this research.

The superiority of the T3 treatment is also reflected in its balance between growth stimulation and conservation of nutritive value. This formulation promoted the highest levels of protein, fat and fiber in all evaluations. The joint action of organic nutrients and beneficial microorganisms present in the biol seems to have enhanced root activity and nutrient uptake efficiency, in agreement with that described by Gil-Ramírez et al. (2023). In addition to the observed values, the linear regression models presented in Table 4 show a robust fit to the experimental data. The crude protein and crude fat variables showed negative slopes, reflecting a decrease with increasing forage age. In contrast, crude fiber, ash and dry matter exhibited increasing trends. The coefficients of determination (R^2) were high for all the variables analyzed, with protein (0.999), fat (0.987), fiber (0.993) and dry matter (0.993) standing out. Ash (0.931), which supports the reliability of the results and the consistency of the observed effects.

Table 4. Linear regression of bromatological variables.

Variable	Model	R^2	p
PB	$y = -0.0708x + 34.375$	0,999	p<0.05
GB	$y = -0.0169x + 9.4483$	0,987	p<0.05
FB	$y = 0.08121x + 16.106$	0,993	p<0.05
CT	$y = 0.0353x + 11.224$	0,931	p<0.05
MS	$y = 0.0576x + 18.669$	0,909	p<0.05

CONCLUSIONS

The research shows that the application of biol at different doses has a positive and significant effect on the morphological and bromatological variables of *Brachiaria decumbens* in the Ecuadorian Amazon. In particular, the maximum dose (T3) proved to be the most efficient in promoting vigorous growth, reaching greater height, greater number of stems and leaves, as well as a strengthened stem diameter, which reflects an optimized structural development. In addition, T3 led to a notable improvement in the nutritional quality of the forage, evidenced by increases in crude protein, fat, fiber and dry matter, aspects traceable to the action of beneficial microorganisms present in the biool that facilitate the solubilization and absorption of nutrients.

These results indicate that the use of biol not only increases pasture productivity, but also improves its nutritional value, contributing to a more balanced diet for livestock in regions with poor and eroded soils. The sustainability and environmental compatibility of the biofertilizer reinforce its potential as a viable alternative for responsible agricultural practices.

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