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Analysis of biomass yield and chemical composition of elephant grass at different growth stages

Análise da produção de biomassa e composição química do capim elefante em diferentes estágios de crescimento

Análisis de la producción de biomasa y la composición química del pasto elefante en diferentes etapas de crecimiento

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Abstract
Planting conditions and harvest timing significantly influence the morphological composition and quality of elephant grass. The objective of this study was to evaluate the dry matter production and compositional changes of elephant grass in terms of ash, extractives, protein, lignin, cellulose, and hemicellulose contents at different growth stages (60 days, 90 days, 120 days, and 60 days regrowth) to recommend ideal harvest periods for specific applications. During the experimental period, favorable climatic conditions prevailed, with temperatures ranging from 17.3 °C to 32.2 °C and cumulative precipitation totaling 1,178 mm. Ash content decreased from 7.2% at 60 days to 4.8% at 120 days, while lignin content increased significantly from 16.9% to 32.7%, reflecting structural changes with plant maturity. Based on these findings, shorter harvest intervals (e.g., 60 days or 60-day regrowth) are recommended for energy generation and forage production due to the lower ash content and higher biodegradability. In contrast, longer growth periods (e.g., 90-120 days) are preferable for applications requiring higher cellulose, hemicellulose, and lignin fractions in lignocellulosic biorefineries.
Keywords: Leaves; Stems; Cellulose; Hemicellulose; Lignin.

Resumo
As condições de plantio e o momento da colheita influenciam significativamente a composição morfológica e a qualidade do capim-elefante. O objetivo deste estudo foi avaliar a produção de matéria seca e as mudanças composicionais do capim elefante em termos do teor de cinzas, extrativos, proteína, lignina, celulose e hemicelulose em diferentes estágios de crescimento (60 dias, 90 dias, 120 dias e 60 dias de rebrote) para recomendar períodos ideais

de colheita para aplicações específicas. Durante o período experimental, as condições climáticas favoráveis prevaleceram, com temperaturas variando de 17,3 °C a 32,2 °C e a precipitação acumulada totalizando 1.178 mm. O teor de cinzas diminuiu de 7,2% em 60 dias para 4,8% em 120 dias, enquanto o teor de lignina aumentou significativamente de 16,9% para 32,7%, refletindo mudanças estruturais com a maturidade da planta. Com base nessas descobertas, intervalos de colheita mais curtos (por exemplo, 60 dias ou 60 dias de rebrote) são recomendados para geração de energia e produção de forragem devido ao menor teor de cinzas e maior biodegradabilidade. Em contraste, períodos de crescimento mais longos (por exemplo, 90-120 dias) são preferíveis para aplicações que exigem maiores frações de celulose, hemicelulose e lignina em biorrefinarias lignocelulósicas.
Palavras-chave: Folha; Caule; Celulose; Hemicelulose; Lignina.

Resumen
Las condiciones de siembra y el momento de la cosecha influyen significativamente en la composición morfológica y la calidad del pasto elefante. El objetivo de este estudio fue evaluar la producción de materia seca y los cambios en la composición del pasto elefante en términos de contenido de cenizas, extractivos, proteínas, lignina, celulosa y hemicelulosa en diferentes etapas de crecimiento (60 días, 90 días, 120 días y 60 días de rebrote) con el fin de recomendar períodos de cosecha ideales para aplicaciones específicas. Durante el período experimental, prevalecieron condiciones climáticas favorables, con temperaturas que oscilaron entre 17.3 °C y 32.2 °C, y una precipitación acumulada de 1,178 mm. El contenido de cenizas disminuyó del 7.2% a los 60 días al 4.8% a los 120 días, mientras que el contenido de lignina aumentó significativamente del 16.9% al 32.7%, reflejando cambios estructurales asociados a la madurez de la planta. Con base en estos hallazgos, se recomiendan intervalos de cosecha más cortos (por ejemplo, 60 días o rebrote de 60 días) para la generación de energía y la producción de forraje debido al menor contenido de cenizas y mayor biodegradabilidad. En contraste, los períodos de crecimiento más prolongados (por ejemplo, 90–120 días) son más adecuados para aplicaciones que requieren mayores fracciones de celulosa, hemicelulosa y lignina en biorrefinerías lignocelulósicas.
Palabras clave: Hojas; Tallos; Celulosa; Hemicelulosa; Lignina.

1. Introduction

Elephant grass (*Pennisetum purpureum*), belonging to the Poaceae family, has gained significant attention in recent years due to its exceptional productivity (Johannes et al., 2024; Dresch et al., 2025). As a C4 grass, it demonstrates remarkable carbon fixation efficiency, sequestering approximately 40 tons of carbon dioxide per hectare annually (Johannes et al., 2024). This efficiency contributes to its impressive dry matter yields, which range from 5 to 47 Mg ha^{-1} $year^{-1}$, depending on cultivation and management practices (Fedenko et al., 2013). The capacity to sequester atmospheric carbon dioxide and produce substantial dry matter makes elephant grass a highly promising candidate for both forage applications and as a feedstock for high-value-added products such as biochar, ethanol, xylitol, and nanocellulose (Ferreira et al., 2019; Flores et al., 2012; Vargas et al., 2023; Yuan et al., 2024; Dresch et al., 2025). This potential is particularly relevant as the demand for alternative raw materials grows, driven by the need to identify crops with high conversion potential to complement or substitute staples like maize and sugarcane during periods of reduced yield (Boschiero et al., 2023).

In addition to its high productivity, elephant grass has garnered attention for its robust growth and low resource requirements. It thrives across a wide range of climates, exhibits strong drought resistance, and achieves high yields even in marginal soils, making it an ideal candidate for sustainable biomass energy production. Furthermore, its role in land management and erosion control enhances its ecological value (Johannes et al., 2024). With rapid growth and suitability for repeated harvesting, elephant grass presents an economically viable option for farmers in both developed and developing countries.

The composition of elephant grass is strongly influenced by both the plant's age and seasonal conditions. As the plant matures, its age directly impacts its suitability for forage and energy production due to significant changes in its chemical composition, particularly between the green leaves and the stems. Green leaves contain higher hemicellulose content and lower levels of cellulose and lignin compared to the stems. In contrast, lignin content, which serves as a barrier to digestibility, increases with age, especially in stems (Rueda et al., 2020). Despite these known changes, few studies have examined the compositional variations in elephant grass as it ages. However, investigating the chemical composition of elephant grass biomass is essential for evaluating its energy potential and identifying its viability for industrial applications.

Therefore, the objective of this study was to analyze the dry matter production of elephant grass and evaluate compositional changes at different growth stages (60 days, 90 days, 120 days, and 60-day regrowth). The goal was to identify optimal growth periods for various applications, maximizing the value of elephant grass and promoting both sustainability and economic benefits.

2. Methodology

This study was based on an experimental research design with a mixed approach, conducted partly in the field and partly in the laboratory, with a predominantly quantitative nature (Pereira et al., 2018). Data analysis included descriptive statistics using bar charts, means, standard deviations, absolute frequencies, and relative percentages. In addition, inferential statistical tests were applied to assess the significance of the results (Vieira, 2021).

2.1 Biomass planting

Elephant grass (*Pennisetum purpureum*) was planted in October 2023 in the experimental area at the Federal University of Fronteira Sul, *Campus* Chapecó, SC, Brazil (-27.119452, -52.706157). The experimental unit measured 4 m in length and 0.8 m in width, covering a total area of 3.2 m². Before planting, the area was prepared by harrowing and subsoiling to a depth of 15 cm, with furrows spaced 80 cm apart. The grass was planted at a density of 16 to 20 nodes per meter within the furrows, using 2 to 3 cuttings per plot. Each cutting was then covered with approximately 10 cm of soil. Weed control was performed 20 days after the grass emerged to optimize growth conditions.

2.2 Biomass harvesting

Elephant grass was manually harvested at different stages of raw material age: 60, 90, and 120 days. After harvesting the biomass at 60 days, a regrowth sample was collected by performing a subsequent cut when the regrown biomass reached 60 days of age (referred to as the '60-day regrowth' sample). Each stage (60, 90, 120 days, and 60-day regrowth) was harvested in quadruplicate on 20th December 2023, 25th January 2024, and 21st February 2024, respectively. During each harvest, an area measuring 1.00 m by 0.20 m was cut 10 cm above ground level in each plot. The harvested biomass was immediately transported to the laboratory, where it was separated into leaves and stems. The separated plant material was weighed and dried in a forced-air circulation oven at 60 °C for 72 hours. After drying, the fractions were reweighed to determine water content, dry matter yield, and the proportion of leaves to stems.

The dried leaves and stems were ground in a Wiley knife mill (Solab model SL–32, Piracicaba, SP, Brazil) and sieved to obtain particles with a maximum diameter of 0.60 mm. The ground material was stored in airtight bags for subsequent physical and chemical characterization analysis.

During the experimental period, precipitation and temperature data (minimum, average, and maximum) were obtained from the Environmental Resources and Hydrometeorology Information Center of Santa Catarina (EPAGRI/CIRAM) (Epagri, 2024).

2.3 Compositional characterization of biomass

The chemical composition of stems and leaves from elephant grass was analyzed at each raw material age in terms of ash, extractives, protein, lignin, cellulose, hemicellulose, acid detergent fiber (ADF), and neutral detergent fiber (NDF). All analyses were performed in quadruplicate.

The ash content was determined using the method described by the National Renewable Energy Laboratory (NREL) (protocol NREL/TP-510-Method 42622) (Sluiter et al., 2008a), with modifications. Briefly, 0.5 g of biomass was calcined at 800 °C for 2 hours in a muffle (Cientec, Model CT-380, Belo Horizonte, MG, Brazil) until a constant weight was achieved.

Extractives were determined following the NREL protocol (NREL/TP-510-42619) (Sluiter et al., 2008b). In this procedure, 2 g of biomass was weighed into an extraction thimble and subjected to sequential extraction with water and ethyl alcohol in a Soxhlet extractor. The extraction process continued until the solvent became colorless, indicating the completion of extraction. After extraction, the thimble was dried in an oven at 105 °C until a constant weight was achieved. The extractive content was calculated based on the weight difference before and after extraction.

Protein content was determined using the Kjeldahl method (N = 6.25), with some modifications (Schmidt et al., 2025). Initially, 0.2 g of biomass, 1 mL of hydrogen peroxide (H_2O_2), 2 mL of sulfuric acid (H_2SO_4), and 0.7 g of digestion mixture (containing 100 g of sodium sulfate [Na_2SO_4]; 10 g of copper sulfate [$CuSO_4$]; and 1 g of selenium) were added to a digestion tube and digested at 350 °C until an emerald green color was achieved (Schmidt et al., 2025). After digestion, the sample was diluted in 25 mL flasks. A 10 mL portion of the diluted sample was neutralized with sodium hydroxide (NaOH) (10 mol/L) and distilled using a nitrogen distiller (Marconi, Model MA-036, Piracicaba, SP, Brazil). The amount of nitrogen in the sample was determined by titration with H_2SO_4 (0.025 mol/L), using boric acid (H_3BO_3) as an indicator. A blank was run without the addition of biomass. The protein content was calculated using Eq. 1.

$$\% \text{ Protein} = [(((V_A - V_B) * 0.7 * 2.5 * 5{,}000)/10{,}000) * 6.25] \quad (1)$$

Where V_A is the volume of H_2SO_4 used in the titration of the sample, and V_B is the volume of H_2SO_4 used in the blank titration.

Cellulose, hemicellulose, and lignin were determined following the method described by NREL (protocol NREL/TP-510-42625) (Sluiter et al., 2012). Briefly, 0.3 g of biomass, free of extractives, was hydrolyzed with 3.0 mL of sulfuric acid (H_2SO_4) (72% v/v) in a water bath at 30 °C for 1 hour. The samples were then diluted to 4% with 84 mL of deionized water, autoclaved at 121 °C for 1 hour, and vacuum filtered. The solid fraction was washed with 500 mL of deionized water and dried in an oven at 105 °C until a constant weight was achieved to determine the acid-insoluble lignin content. The liquid sample was separated into two fractions: one aliquot was used to measure the acid-soluble lignin using a UV-Vis Spectrometer (UV-1800, Nova Instruments, Piracicaba, SP, Brazil), and the other was neutralized with calcium carbonate powder to a pH of 5-6 and filtered through 0.2 µm syringe filters for determination of carbohydrates, acetyl groups, and acid-soluble lignin by High-Performance Liquid Chromatography (HPLC) (Shimadzu, Barueri, SP, Brazil).

Sugars (e.g., glucose, xylose, and arabinose) and inhibitors (e.g., acetic acid, furfural, and 5-hydroxymethylfurfural) were analyzed using an HPLC (LC-MS 2020, Shimadzu, Japan) equipped with a refractive index detector (RID-10A) and an organic acid column (Aminex HPX-87H, Bio-Rad) (Lucaroni et al., 2022; Dresch et al., 2025). For carbohydrate analysis, a mobile phase of 5 mM sulfuric acid at 50 °C with a flow rate of 0.6 mL/min was used. For inhibitor analysis, an SPD-M20A with an NST-18 column was employed, using a mobile phase of 85:15 acetonitrile/water with 1% acetic acid at 40 °C and a flow rate of 0.8 mL/min (Lucaroni et al., 2022; Dresch et al., 2023; Vargas et al., 2023).

Cellulose and hemicellulose content were estimated using Eqs. 2 and 3, respectively.

$$\% \text{ Cellulose} = [((((0.95 * Ce) + (0.90 * G) + (1.29 * HMF) * V) * (1 - (E/100)))/M] \quad (2)$$

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$$\% \text{ Hemicellulose} = [(((0.88 * X) + (1.37 * F) + (0.72 * AA) + (0.88 * ARA) * V) * (1 - (E/100)))/M] \quad (3)$$

Where Ce: Cellobiose (g/L); G: Glucose (g/L); HMF: 5-hydroxymethylfurfural (g/L); V: Filtration volume (0.087 L); E: Extractives (%); M: Biomass (g); X: Xylose (g/L); F: Furfural (g/L); AA: Acetic Acid (g/L); and ARA: Arabinose (g/L).

The percentage content of neutral detergent fiber (NDF) was estimated as the sum of cellulose, hemicellulose, and lignin, while acid detergent fiber (ADF) was calculated as the sum of lignin and cellulose.

2.4 Statistical analysis

The results were analyzed using an analysis of variance (ANOVA) followed by Tukey's test, with a 95% confidence level ($p < 0.05$), using OriginPro 2024b Software (version 10.15).

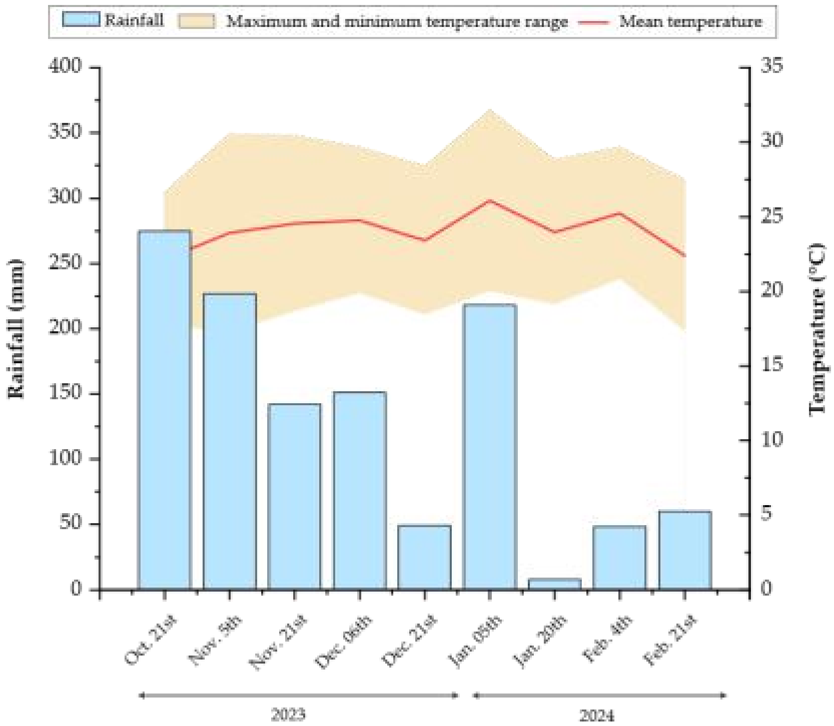
3. Results and Discussion

3.1 Climatic conditions

Climatic conditions play a crucial role in the development of crops, including elephant grass. During the experimental period (October 2023 to February 2024) (Figure 1), the minimum temperatures ranged from 17.29 °C to 20.81 °C, while the maximum temperatures varied between 26.67 °C and 32.20 °C. Although elephant grass thrives in temperatures between 28 °C and 33 °C (Marafon et al., 2021; Pereira et al., 2021), the observed temperatures were slightly below this optimal range; however, temperatures below 15 °C are reported as limiting for the biomass production of elephant grass (Freitas et al., 2017).

Elephant grass typically concentrates 70–80% of its production during rainy seasons (Pereira et al., 2021). Over the four-month experimental period, a significant accumulated precipitation of 1,178 mm was recorded. Although high precipitation rates were observed, the combination of elevated temperatures and intense sunlight during the experimental period provided conditions close to the ideal growth scenario for elephant grass (Johannes et al., 2024; Pereira et al., 2021). It is important, however, to monitor precipitation volumes, particularly in regions with lower temperatures, as elephant grass does not adapt well to waterlogged soils or areas prone to prolonged flooding (Pereira et al., 2021).

Figure 1 - The datasets present the biweekly precipitation and the maximum, average, and minimum temperatures recorded during the experimental period (October 2023 to February 2024) in Chapecó, SC, a southern region of Brazil.



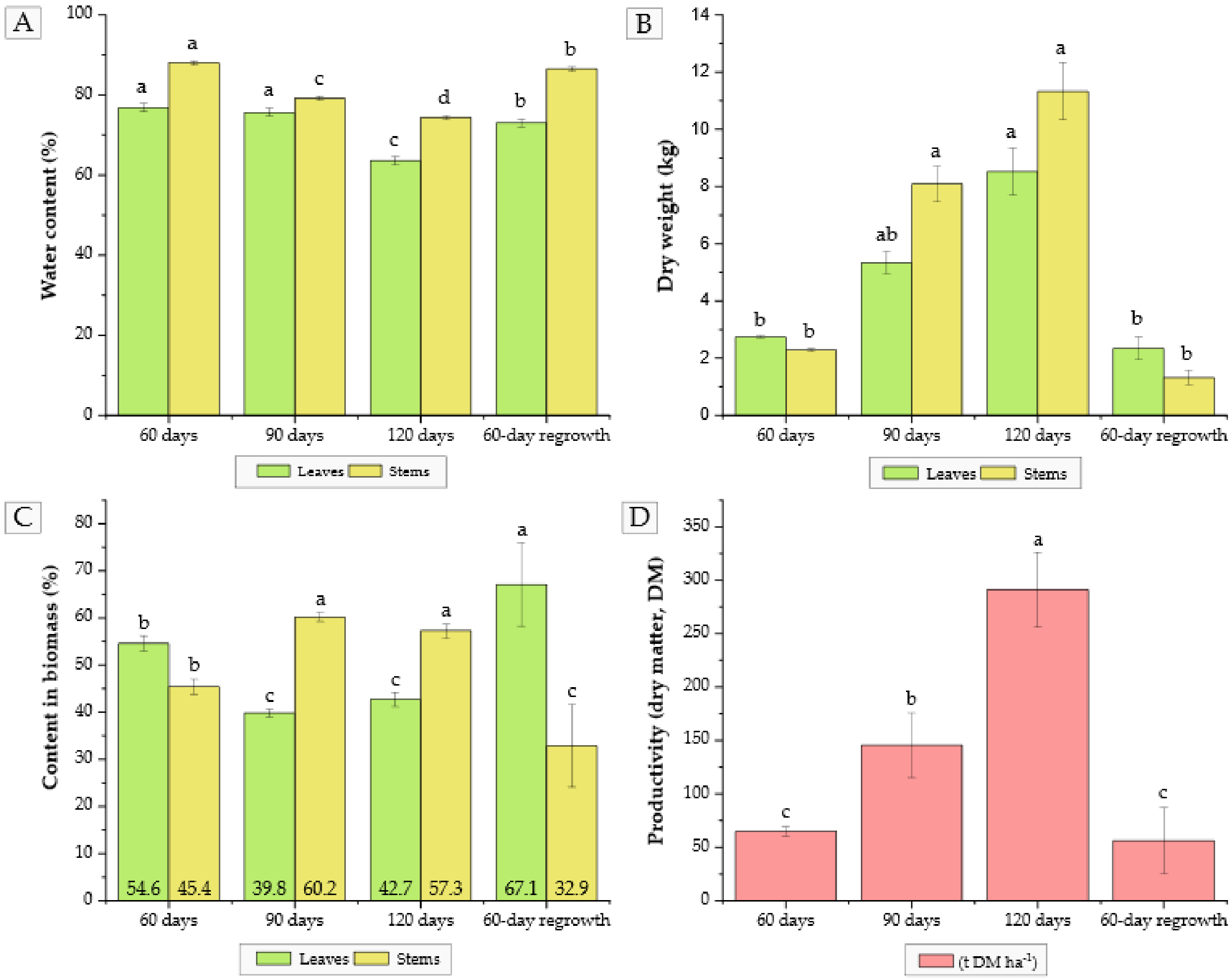
Source: Epagri (2024).

3.2 Biomass yields

Analyzing dry matter yield is crucial for assessing the viability and profitability of biomass for bioenergy applications and biotechnological industries. This is particularly important for biomasses like elephant grass, which is gaining attention as a potential supplement or substitute for traditional raw materials used in these processes. Its increasing prominence is attributed to its adaptability to various soil types and its exceptional productivity.

At all raw material ages, the water content of elephant grass exceeded 65% (Figure 2A), with stems containing slightly more water than leaves. This difference occurs because water is transported from the roots to the leaves through the xylem, a conductive tissue extending throughout the plant (Nobel, 2017). Additionally, the plant's transpiration process contributes to this variation, as water evaporates from the leaves into the atmosphere (Takara & Khanal, 2015). Raw material harvested at 90 and 120 days had lower water content, likely due to higher temperatures or reduced precipitation before harvest. These climatic factors can accelerate water loss through transpiration and limit water uptake from the soil, resulting in reduced water levels in the biomass.

Figure 2 - Water content (A), dry weight (B), leaf-to-stem ratio (C), and productivity (D) of elephant grass at different stages of raw material age. Data represents the mean ± standard error from four replicates. Different letters for each raw material age indicate significant differences according to Tukey's test ($p < 0.05$).



Source: Authors (2025).

As the plant matures, the water content decreases and is replaced by lignin through the lignification process (Yuan et al., 2024). This trend was evident in the present study, which showed a reduction in water content and a corresponding increase in lignin percentage as the plant reached higher maturity levels (Table 1). Lignin plays a vital role in the plant by enhancing rigidity, increasing resistance, and providing protection against pests and diseases (Dresch et al., 2025). In a related study, the water content of elephant grass at 2 months of age exceeded 80%, which is significantly higher compared to the values observed in the plant at more advanced developmental stages (Yuan et al., 2024).

Table 1 - Chemical composition (%) of leaves, stems, and whole plant of elephant grass at different harvest stages. Different letters for each raw material age indicate significant differences according to Tukey's test ($p < 0.05$).

	Chemical composition (%)						
	Ash	**Extractives**	**Lignin**	**Protein**	**Cellulose**	**Hemicellulose**	**Total**
Leaves							
60 days	5.8 ± 0.2^{b}	24.0 ± 0.5^{a}	15.7 ± 0.3^{c}	8.2 ± 0.0^{c}	28.7 ± 1.5^{a}	13.0 ± 0.6^{a}	95.4 ± 3.1
90 days	6.4 ± 0.1^{b}	21.8 ± 0.8^{b}	16.1 ± 1.5^{c}	9.1 ± 0.3^{b}	25.3 ± 2.3^{ab}	14.0 ± 1.5^{a}	92.7 ± 6.3
120 days	6.1 ± 0.6^{b}	20.6 ± 0.3^{bc}	35.0 ± 4.4^{a}	9.8 ± 0.2^{b}	21.1 ± 1.4^{b}	12.7 ± 0.6^{a}	105.4 ± 7.4
Regrowth	7.4 ± 0.2^{a}	20.0 ± 0.2^{c}	23.7 ± 1.6^{b}	12.0 ± 0.6^{a}	25.8 ± 1.0^{a}	14.6 ± 0.4^{a}	103.4 ± 4.0
Stems							
60 days	8.9 ± 0.3^{a}	34.5 ± 0.2^{a}	18.3 ± 0.4^{c}	3.5 ± 0.3^{b}	26.5 ± 0.9^{b}	11.6 ± 0.5^{b}	103.3 ± 2.5
90 days	5.4 ± 0.4^{b}	25.0 ± 0.5^{b}	22.7 ± 0.8^{b}	1.6 ± 0.0^{d}	31.3 ± 1.0^{ab}	14.2 ± 0.8^{ab}	100.2 ± 3.6
120 days	3.8 ± 0.1^{c}	19.3 ± 0.5^{c}	30.9 ± 1.3^{a}	3.2 ± 0.0^{c}	34.3 ± 3.6^{a}	15.6 ± 1.0^{a}	107.1 ± 6.5
Regrowth	5.5 ± 0.1^{b}	24.8 ± 0.2^{b}	22.5 ± 0.4^{b}	6.0 ± 0.3^{a}	28.2 ± 3.1^{ab}	13.4 ± 2.1^{ab}	100.4 ± 5.8
Total (Leaves + Stems)							
60 days	7.2 ± 0.2^{a}	28.7 ± 0.3^{a}	16.9 ± 0.1^{c}	6.1 ± 0.1^{b}	27.7 ± 1.0^{a}	12.4 ± 0.4^{b}	99.0 ± 2.3
90 days	5.8 ± 0.3^{b}	23.4 ± 0.7^{b}	20.1 ± 0.4^{bc}	4.6 ± 0.1^{c}	28.9 ± 1.0^{a}	14.1 ± 0.4^{a}	96.9 ± 3.0
120 days	4.8 ± 0.2^{b}	19.8 ± 0.5^{d}	32.7 ± 2.4^{a}	6.0 ± 0.1^{b}	28.7 ± 1.5^{a}	14.3 ± 0.6^{a}	106.3 ± 5.2
Regrowth	6.6 ± 0.5^{ab}	21.6 ± 0.2^{c}	23.3 ± 0.9^{b}	10.0 ± 0.4^{a}	26.6 ± 1.2^{a}	14.2 ± 0.4^{a}	102.2 ± 3.6

Source: Authors (2025).

Dry matter content is inversely proportional to water content, meaning that as water content decreases, the amount of dry matter increases (Figure 2B) (Yuan et al., 2024). Dry matter, which represents the plant's total mass excluding moisture, contains all its nutrients (Peixoto et al., 2024). Longer growth periods result in higher dry matter content. For example, elephant grass at 120 days of growth produced 11.33 kg of dry matter in the stem and 8.53 kg in the leaves. In contrast, shorter growth periods, such as 60 days and 60-day regrowth, yielded significantly less dry matter: 2.28 kg and 1.31 kg for the stem, and 2.74 kg and 2.34 kg for the leaves, respectively. These results correspond to the higher water content observed during shorter growth periods, such as 60 days (88.04% for the stem and 76.83% for the leaves) and 60-day regrowth (86.53% for the stem and 73.02% for the leaves). This indicates that as water content decreases, lignin accumulation through lignification increases, leading to a higher percentage of dry matter. Previous studies have also shown that extending the harvest interval for elephant grass leads to increased dry matter content (Freitas et al., 2017). This is significant because dry matter accumulation is a key factor in determining biomass quality, which depends on nutrient density and structural composition (Johannes et al., 2024).

The proportion of leaves and stems in elephant grass directly influences its degradability potential, depending on the intended biomass application (Peixoto et al., 2024). For shorter growth periods, the leaf proportion was higher, reaching 54.6% at 60 days and 67.1% for 60-day regrowth. At 90 and 120 days, the leaf proportion stabilized (Figure 2C). Similarly, a previous study reported a decline in leaf content from 56% to 19% during a 7-month growth period (Na et al., 2016). In contrast, stem content increased from 45.4% at 60 days to 60.2% at 90 days, with no further changes observed between 90 and 120 days. To maximize biodegradability, cutting intervals shorter than 70 days can prevent excessive stem accumulation (Rueda et al., 2020). For example, in this study, the 60-day regrowth yielded a low stem proportion of 32.9%, favoring higher degradability. Reducing the stem proportion decreases the lignified fiber content in the biomass, which enhances biodegradability (Peixoto et al., 2024; Rueda et al., 2020). This adjustment is particularly beneficial for applications requiring lower fiber density and higher digestibility, such as bioenergy production and animal forage.

Biomass productivity is a key indicator for evaluating the potential of biomass as a raw material for high-value products and determining its profitability (Freitas et al., 2017). In this study, elephant grass demonstrated high productivity in terms of dry matter: 64.77 t ha^{-1} at 2 months, 145.48 t ha^{-1} at 3 months, 291.32 t ha^{-1} at 4 months, and 56.35 t ha^{-1} with 2-month Regrowth (Figure 2D). Similar high productivity was reported by Rengsirikul et al. (2011), who found 50.8 t ha^{-1} at 2 months. In contrast, the "mole de volta grande" genotype achieved a much lower productivity of 6.81 t ha^{-1} at the same growth stage (Freitas et al., 2017). For the 3-month growth period, productivity estimates varied, with Rengsirikul et al. (2011) reporting 61.4 t ha^{-1} and Freitas et al. (2018) observing 26.54 t ha^{-1} for the "kind grass" genotype. The productivity in the current study was slightly higher; however, it is important to note that factors such as genotype, cultivation methods, soil conditions, and climate can influence results. The fast growth cycle of elephant grass allows for multiple harvests per year — typically 3 to 4 — and its high productivity makes it a promising option for biofuel production and other high-value economic products (Johannes et al., 2024).

3.3 Chemical composition

The physicochemical composition of biomass can vary depending on factors such as location, growth conditions, harvest time, and the methods used for plant characterization (Johannes et al., 2024). Understanding how the lignocellulosic composition of materials like elephant grass changes is essential for optimizing biomass conversion into high-value products. As mentioned above, the plant’s growth period significantly influences its degree of degradability (Yuan et al., 2024).

The concentration of ash in biomass directly affects its suitability for combustion processes in energy generation. High ash content can obstruct combustion by causing buildups (incrustations) and wear (erosion) in combustion equipment, while lower ash concentrations enhance biofuel production efficiency during pyrolysis (Johannes et al., 2024). In this study, the ash content was relatively low, with the highest concentration found in the stem at the 60-day regrowth period (8.90%), and the lowest in the stem at the 120-day growth period (3.80%) (Table 1). This increase is due to the plant's defense mechanisms, which include both passive (e.g., accumulation of minerals) and active (e.g., biochemical responses) processes in Poaceae family grasses like elephant grass (Takara & Khanal, 2015). A study conducted in Hawaii also supported this trend, showing that elephant grass harvested at 2 months had an ash content of 15.3%, significantly higher than the 8.7% observed in the plant at 8 months of maturity (Takara & Khanal, 2015).

When analyzing the ash content in the entire elephant grass plant, including both leaves and stems, the 120-day growth period showed an ash content of 4.80% (Table 1). This value is lower than those reported by Rengsirikul et al. (2011) (8.80%) and Lee et al. (2010) (9.68%) for elephant grass harvested at 3 months, as well as lower than the value found by Ferreira et al. (2019) for 4-month-old plants (8,10%). The ash content of elephant grass varies significantly, ranging from 3.0% to 16% (Johannes et al., 2024; Takara & Khanal, 2015). A study on 18 varieties found an average ash content of 4.74% (Marafon et al., 2021), while other reports show values of 3.50% (Vargas et al., 2023), 4.90% (Toscan et al., 2022), and 5.70% (Scopel et al., 2023).

Generally, younger plants tend to have higher ash content, with values varying based on the plant's maturity. For biomass intended for combustion, low ash content is preferred, with values below 9% considered optimal (Rengsirikul et al., 2011). High ash concentrations can lead to issues like incrustations and erosion during combustion, but they also indicate the presence of minerals such as potassium, chlorine, and silicon, which can catalyze pyrolysis reactions (Johannes et al., 2024). However, for applications such as activated carbon production, lower ash content (< 3.0%) is more desirable (Johannes et al., 2024). Additionally, the reduction in ash content with plant maturity is closely linked to a decrease in the proportion of leaves. Longer harvest intervals, associated with senescence and the translocation of constituents, have been found to reduce ash concentration (Na et al., 2016). Inorganic components, which make up the "ash," cannot be converted into energy and can reduce biomass quality, especially for processes like fast pyrolysis (Na et al., 2016).

Extractives, though not directly relevant for bioenergy generation, consist of water- and ethanol-soluble components that have valuable applications in food, pharmaceuticals, and the cosmetics industry (Scopel et al., 2020; Scopel & Rezende, 2021). These components can potentially replace products derived from the petroleum industry. The higher concentration of extractives was found in the stems (19.3% to 34.5%, Table 1), and can be attributed to their role in the plant's defense system, protecting it from attacks by microorganisms and insects (Boschiero et al., 2023). Conversely, extractives are less abundant in dried leaves, as they are no longer part of the living plant and no longer require such a defense mechanism (Boschiero et al., 2023). In this study, the extractive content of elephant grass ranged from 19.80% to 28.70%, consistent with the values reported by Scopel et al. (2023) and Vargas et al. (2023), of 20.20% and 17.86%, respectively.

Elephant grass is not known for having high protein content. The highest protein levels were observed in the 60-day regrowth, with values of 6.00% ± 0.30 for the stem, 12.00% ± 0.60 for the leaves, and 10.00% ± 0.40 for the whole biomass (stem + leaves) (Table 1). A study conducted by Yuan et al. (2024) reported protein levels of 14.70% for the stem at 2 months of plant age, significantly higher than the value obtained in the present study (6.00%). The observed differences may be attributed to variations in experimental conditions or plant genotypes.

The holocellulose fraction (cellulose and hemicellulose) and lignin are the primary components of lignocellulosic matrices, accounting for over 60% of the plant's total composition (Takara & Khanal, 2015). Quantifying these constituents is crucial for optimizing biomass treatment processes and its conversion into high-value products. Studies have shown that the plant's growth stage significantly affects the percentage of these components (Yuan et al., 2024), largely due to the lignification process.

In this study, the cellulose content ranged from 21.1% to 28.7% in leaves, 26.5% to 34.3% in stems, and 26.6% to 28.9% in the whole elephant grass plant. No significant differences were observed in cellulose content across the different maturation stages. Similarly, Takara & Khanal. (2015) reported no significant variation in cellulose content across growth stages (2, 4, 6, and 8 months) of elephant grass. Literature values for cellulose content range from 7.18% to 44.70%, while the hemicellulose values range from 3.04% to 38.10% (Table 2). Hemicellulose content in this study was 12.4% (60 days), 14.1% (90 days), 14.3% (120 days), and 14.2% (60-day regrowth), with minimal variation across later growth stages. While holocellulose is less critical in direct combustion due to its low thermal stability and energy activation (Marafon et al., 2021), cellulose and hemicellulose have significant potential for diverse applications. They are used in producing bioplastics, biofilms, composite materials, chemicals, xylitol, xylooligosaccharides, and furfural, for example (Schmidt et al., 2023; Vargas et al., 2023; Woiciechowski et al., 2020; Dresch et al., 2025).

Lignin has traditionally been viewed as a barrier in the enzymatic production of biofuels from cellulose and hemicellulose and as a limiting factor for forage use due to its low digestibility (Johannes et al., 2024; Na et al., 2016). However, recent studies highlight its potential in producing biotechnological products, such as biomaterials, bioplastics, and products for the cosmetic and pharmaceutical industries, particularly due to its antioxidant properties (Ivanova et al., 2023; Tanganini et al., 2024; Xu et al., 2020; Dresch et al., 2025). In this study, lignin content was 16.9% (60 days), 20.1% (90 days), 32.7% (120 days), and 23.3% (60-day regrowth) (Table 1). These values align with the normal range for grasses, reported to vary between 6.70% and 26% (Table 2). Lignin content increased with plant maturity, reflecting the lignification process that enhances cell wall rigidity, microbial resistance, and impermeability (An et al., 2019; Johannes et al., 2024; Lang & Li, 2022).

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However, optimal harvesting times should be aligned with the intended biomass application. Higher lignin content in later stages of plant development may be disadvantageous for forage or bioenergy production. Conversely, later harvesting is more suitable when valorizing cellulose, hemicellulose, and lignin fractions separately for biotechnological applications.

Table 2 - Cellulose, hemicellulose, and lignin values in other studies found in the literature for elephant grass biomass.

Location	Plant Content	Raw Material Age	Content (%)			Reference
			Cellulose	Hemicellulose	Lignin	
Brazil	Stems	2 months	7.18	3.04	7.22	(Yuan et al., 2024)
Thailand	Leaves + Stems	2 months	41.30	25.30	6.70	(Rengsirikul et al., 2011)
Hawaii	Leaves + Stems	2 months	35.00	19.20	15.30	(Takara & Khanal, 2015)
Thailand	Leaves + Stems	3 months	43.90	22.20	10.20	(Rengsirikul et al., 2011)
Hawaii	Leaves + Stems	4 months	38.50	22.80	17.90	(Takara & Khanal, 2015)
Brazil	Leaves + Stems	6 months	36.00	30.30	8.80	(Marafon et al., 2021)
Brazil	Stems	6 months	13.24	~4.10	13.22	(Yuan et al., 2024)
Thailand	Leaves + Stems	6 months	44.70	23.60	12.50	(Rengsirikul et al., 2011)
Hawaii	Leaves + Stems	6 months	38.60	23.40	17.50	(Takara & Khanal, 2015)
Brazil	Stems	8 months	27.39	4.51	13.80	(Yuan et al., 2024)
Brazil	Stems	10 months	~23.00	~4.00	14.35	(Yuan et al., 2024)
Brazil	Leaves + Stems	-	36.00[a]	30.30[a]	8.80[a]	(Marafon et al., 2021)
Brazil	Leaves + Stems	-	35.69	15.26	18.03	(Vargas et al., 2023)
Brazil	Leaves + Stems	-	30.00	24.00	22.50	(Scopel et al., 2023)
Brazil	Leaves + Stems	-	34.50	38.10	13.50	(Toscan et al., 2022)
Brazil	Leaves + Stems	-	34.30	19.60	25.80	(Dresch et al., 2025)

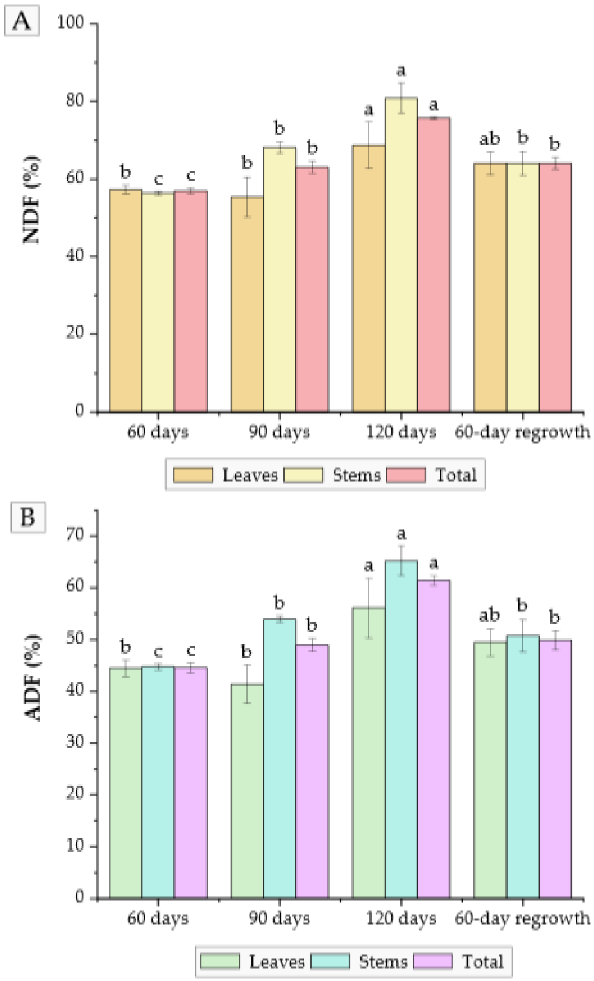
[a] The mean value of 18 different genotypes of elephant grass. Source: Authors (2025).

Neutral detergent fiber (NDF) and acid detergent fiber (ADF) are critical indicators for assessing lignocellulosic biomass quality (Yuan et al., 2024). NDF encompasses the indigestible fibrous components of biomass — cellulose, hemicellulose, and lignin — and is significantly affected by harvest timing (Peixoto et al., 2024). NDF serves as a key indicator of lignocellulosic biomass quality since the cell wall is a primary energy source for ruminant microorganisms. However, elevated NDF levels can restrict microbial access to carbohydrates, posing challenges similar to those encountered in ethanol production (Na et al., 2016). In contrast, ADF refers to the fiber fraction comprising cellulose and lignin and is closely linked to forage digestibility and energy content. Forages with lower ADF values generally provide higher energy, making them more suitable for ruminant diets (Fahey et al., 2019; Yuan et al., 2024).

Figure 3a illustrates that elephant grass with a 120-day growth period exhibited the highest NDF percentages (80.80% for the stem, 68.80% for the leaf, and 75.70% for the whole biomass), showing a significant difference compared to all other harvest times, except for the 60-day regrowth leaf (64.10%). Similar NDF values, ranging from 75.90% to 78.65%, have been reported in other studies for elephant grass harvested at 4 months of growth (Freitas et al., 2017), closely aligning with the findings of this study. The shortest growth period, 60 days, had the lowest NDF value at 65.20%, which is consistent with the values reported for elephant grass at 2 months of growth in the literature (68.03%–71.37%) (Freitas et al., 2017). As shown in Figure 3A, the NDF percentage increases with the plant's development, primarily due to the lignification process. Similarly, a study by Yuan et al. (2024) observed a rise in NDF values as the growth period extended from 2 months (44.99%) to 8 months (69.08%).

As previously aforementioned, understanding the NDF content in elephant grass is crucial for assessing forage quality. While a high NDF percentage indicates increased fiber content, it also results in reduced digestibility in the gastrointestinal tract, slowing food digestion in ruminants and delaying feed intake (Peixoto et al., 2024). Therefore, for optimal silage production, elephant grass should be harvested between 35 and 45 days of growth (Peixoto et al., 2024), as this period provides the best balance of degradability and nutritional quality. Other studies suggest that harvesting up to 3 months of growth is ideal for cattle feed (Rengsirikul et al., 2011). Longer growth periods lead to higher fiber content and lower nutrient levels, making the grass less suitable as animal forage (Yuan et al., 2024). In contrast, for the valorization of elephant grass as a raw material for producing high-value-added products such as biofuels, bioplastics, and phenolic compounds, longer growth periods are more advantageous.

Figure 3 - Neutral detergent fiber (NDF) (A) and acid detergent fiber (ADF) (B) content of elephant grass at different stages of raw material age. Data represents the mean ± standard error from four replicates. Different letters for each raw material age indicate significant differences according to Tukey's test ($p < 0.05$).



Source: Authors (2025).

Regarding the ADF values, a clear upward trend was observed as the plant matured. At 60 days, the ADF content was 44.60%, increasing to 61.40% at 120 days (Figure 4B). The ADF value for elephant grass at 2 months of growth was consistent with those reported in other studies (37.22–39.23%) (Freitas et al., 2017). However, the ADF content for the 120-day growth period exceeded the range reported in other studies (43.26–46.82%) (Freitas et al., 2017). As with NDF, elevated ADF levels make the plant less suitable for use as animal forage, as they are associated with lower digestibility and reduced energy availability.

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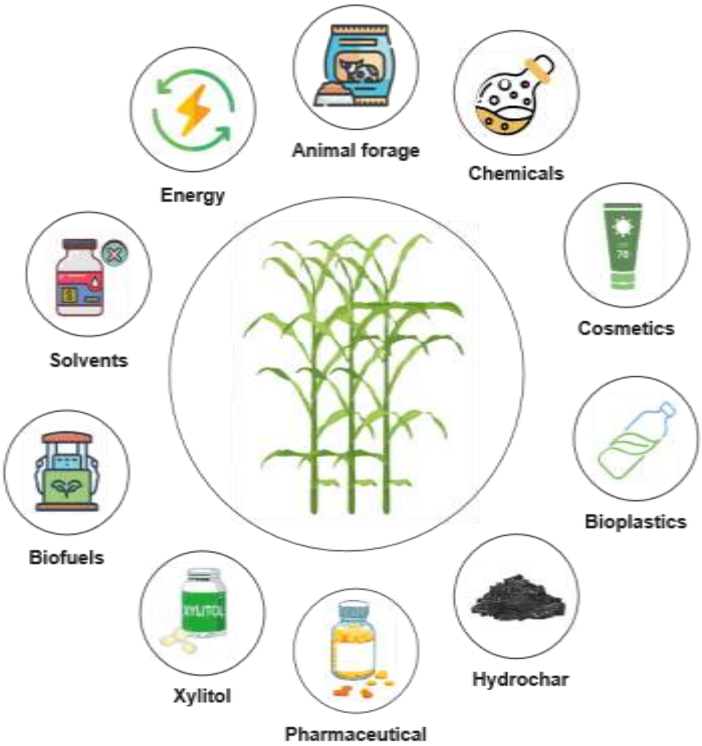
3.4 Overview of potential applications of elephant grass

As evidenced by the characterization results in this study, the composition of elephant grass varies significantly with its growth stage. Therefore, determining the optimal growth period is crucial and should align with the intended final application. Despite its wide range of applications, elephant grass is still primarily utilized as forage (Figure 4) (Peixoto et al., 2024; Pereira et al., 2021). For this purpose, harvesting within a growth period of no more than 3 months is recommended, as it enhances digestibility and minimizes the lignification level of the biomass.

Figure 4 - Overview of potential applications of elephant grass biomass.



Source: Authors (2025).

Another prominent application of elephant grass is in the production of bioenergy and biofuels (Sousa et al., 2016; Fedenko et al., 2013; Iyyappan et al., 2023; Scopel et al., 2020; Takara & Khanal, 2015; Vargas et al., 2023; Dresch et al., 2025). For direct biomass combustion to generate energy, a lower ash content is preferable, which makes longer growth periods more desirable. In contrast, for biofuel production via cellulose conversion into glucose and subsequently ethanol, lignin content plays a critical role. High lignin levels impede enzyme access to cellulose during the saccharification process. Therefore, as this study found no significant differences in cellulose content across growth stages, biomass harvested at shorter growth periods would be more suitable due to its lower lignin content.

However, in recent years, the concept of biorefinery has gained significant attention, focusing on minimizing waste and enhancing process sustainability. This approach seeks to valorize all biomass fractions — extractives, lignin, cellulose, and hemicellulose — for the production of various high-value-added products (Boschiero et al., 2023; Sousa et al., 2016; Ferreira et al., 2019; Iyyappan et al., 2023; Pereira et al., 2021; Scopel et al., 2020; Vargas et al., 2023; Yuan et al., 2024; Dresch et al., 2025), as illustrated in Figure 4. To support this objective, selecting a growth period that maximizes the yield of these components is critical. For example, growth periods of 90 or 120 days may be optimal for balancing lignocellulosic fractions and enhancing their potential applications.

4. Conclusion

Elephant grass is a widely cultivated species, valued for its ease of cultivation and high dry matter productivity. However, its lignocellulosic composition varies with the plant's developmental stage, making it essential to understand these changes over time to optimize subsequent processing. Among the parameters quantified in this study (ash, extractives, protein, cellulose, hemicellulose, lignin, acid detergent fiber, and neutral detergent fiber), cellulose was the only component that showed no significant variation across the four tested harvesting times (60 days, 90 days, 120 days, and 60-day regrowth). These findings provide valuable insights into structural modifications in elephant grass, aiding in selecting optimal harvest times based on specific applications. For forage or energy production, shorter harvest periods are recommended to minimize lignin content and enhance biodegradability. In contrast, longer growth periods are preferable when higher dry matter yields and the simultaneous valorization of cellulose, hemicellulose, and lignin fractions are desired.

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Credit Authorship Contribution Statement

Aline Perin Dresch: Conceptualization; Methodology; Investigation; Data curation; Formal Analysis; Visualization; Writing – original draft; Writing – review & editing; Resources. *Kaillany Eduarda Gonçalves Lipes:* Investigation; Writing – original draft. *Siumar Pedro Tironi:* Methodology; Investigation. *Odinei Fogolari:* formal analysis. *Guilherme Martinez Mibielli:* Conceptualization; Investigation; Supervision; Resources. *João Paulo Bender:* Conceptualization; Investigation; Supervision; Resources; Writing – review & editing. *Joel Gustavo Teleken:* Resources; Writing – review & editing.

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