

Article

Does Carbon-Based Material Influence Tropical Grasses Under Water Deficit?

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Highlights

What are the main findings?

- Water deficit reduced the growth and forage production of Sabiá grass, whereas Cayana grass showed greater productive stability under the water-deficit regime.
- Carbon-based material increased forage mass in Sabiá grass regardless of water availability and mitigated the effect of water deficit on phyllochron, maintaining leaf emergence under water-limited conditions.

What are the implications of the main findings?

- The effectiveness of carbon-based material may depend on forage grass genotype and the conditions under which water deficit occurs.
- Carbon-based material may be a promising complementary technology to improve productivity stability of drought-sensitive tropical grasses under irregular water availability.

Abstract

Carbon-based materials (CM) have been investigated for their potential to improve plant responses to abiotic stresses, including water deficit, but their effects on tropical forage grasses remain poorly understood. The objective of this study was to assess whether CM affects the productivity of tropical grasses under water deficit. Two experiments were conducted, each with cultivars that are hybrids of *Urochloa* spp.: Sabiá grass and Cayana grass. The study was carried out in a greenhouse using four treatments arranged in a 2 × 2 factorial design. Treatments consisted of two water availability conditions (with and without water deficit) and two strategies (with and without application of CM). Evaluations were performed over two growth cycles. For Cayana grass, forage production was not affected by water deficit, and CM did not increase forage production. In contrast, water deficit impaired the development of Sabiá grass, and CM application increased forage mass regardless of water availability and maintained a lower phyllochron under water deficit. Carbon-based material increased forage production in Sabiá grass but not in Cayana grass,



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indicating that their effects may depend on the grass genotype and the conditions under which water deficit occurs.

Keywords: carbon-based materials; drought tolerance; pasture; water stress

1. Introduction

Carbon dots (CDs), a class of carbon-based materials, have recently gained increasing attention, with a growing number of publications [1] highlighting not only their direct physiological effects on plants but also their potential use in the formulation of agrochemicals. Their versatility allows them to be integrated into fertilizers and plant growth promoters, improving nutrient delivery and stress resilience, which aligns with the demand for more sustainable and efficient agricultural inputs [2].

In addition, carbon dots stand out as low-cost nanomaterials derived from abundant and renewable sources, offering both economic and environmental advantages. Their biocompatibility, low toxicity, and high stability in aqueous media have driven increasing research into agricultural applications [2,3], where they can contribute to improved resource-use efficiency and sustainable production systems [4].

One of the most relevant properties of carbon dots is their ability to convert ultraviolet radiation into visible light [5], thereby increasing photon availability for chloroplasts and enhancing photosynthetic efficiency [6]. This characteristic provides opportunities to improve net photosynthesis, which may promote plant development and resilience.

Studies in different crops have demonstrated positive effects, including increases in photosynthetic efficiency, root and shoot growth, phytohormone regulation, and nutrient uptake. In rice, yield improvements of up to 14% have been reported, along with increased disease resistance [7]. In maize, the combined use of nitrogen and carbon dots enhanced productivity by up to 24% [8].

Under water stress conditions, the application of carbon dots in soybean increased ground cover, leaf area, chlorophyll content, maximum efficiency of photosystem II, and relative electron transport rate. These changes led to higher leaf water content, greater osmolyte accumulation, enhanced antioxidant activity, and approximately 25% higher grain yield [9]. Despite these promising results, caution is required regarding concentrations, since some carbon nanomaterials have shown phytotoxic effects at elevated doses [10]. Therefore, further research is needed to optimize carbon dot concentrations for different crop species [11].

Several species of the introduced tropical grass genus *Urochloa* are widely used in pastures for livestock production in Brazil and worldwide. One of the main limitations of these systems is the strong seasonality of forage production, driven primarily by irregular rainfall distribution. During dry seasons, forage supply is drastically reduced, which compromises stocking capacity, animal performance, and farm profitability. Some species of *Urochloa* spp. (syn. *Brachiaria* spp.), such as *U. brizantha*, *U. decumbens*, and *U. mutica*, have shown tolerance to water deficit for up to 14 days, while *U. humidicola* and *U. dictyoneura* have shown tolerance for up to 28 days [12]. Nevertheless, the literature lacks information about the water deficit tolerance of hybrids derived from *Urochloa* species, including the cultivars Sabiá and Cayana.

The Cayana grass is a hybrid of *Urochloa ruziziensis* and *Urochloa brizantha*. A study testing the tolerance of Cayana grass to different water availabilities showed a low tolerance, with reductions of 75% in forage mass production and 90% in root mass [13]. The Sabiá grass is a hybrid of *Urochloa ruziziensis* and *Urochloa brizantha*, developed by Barenbrug and

marketed as a cultivar suitable for grazing systems, yielding from 9.1 to 28.7 ton/ha/year of dry forage mass, and more tolerant to the dry season [14].

Technological alternatives capable of increasing the resilience of pastures to water deficit are therefore essential to improve productivity without expanding agricultural areas. Although several studies have evaluated carbon dots in grain crops, there is a lack of information about the effects of carbon-based material (CM) on tropical forage grasses used in grazing systems. This gap is particularly relevant, as perennial grass differs from annual crops in terms of growth dynamics, perenniality, and the capacity of their root systems to act as reservoirs of organic compounds and nutrients during stressful periods.

Thus, evaluating the effects of CM on tropical forage grasses represents an innovative approach with the potential to increase forage accumulation and improve stocking rates. These responses may contribute to the sustainability of livestock production systems in regions subject to climatic variability. In addition, this approach may help mitigate the impacts of climatic phenomena such as El Niño and La Niña, which can alter water availability and intensify its variability.

Therefore, the objective of this study was to determine whether carbon-based material affects the productivity of two tropical forage grasses used in grazing systems under non-deficit moisture with irrigation and simulated water-deficit conditions.

2. Materials and Methods

2.1. Local and Experimental Design

Two experiments were conducted simultaneously in the same greenhouse at the Federal University of Rondonópolis, Rondonópolis, Mato Grosso, Brazil (16°27'49" S, 54°34'4" W; approximately 227 m altitude). The experimental units of both grasses were placed on the same greenhouse bench throughout the experimental period and were therefore exposed to the same general greenhouse environmental conditions. According to the Köppen classification, the climate in the region is humid tropical (Aw). The experiments were conducted in a completely randomized design in a 2 × 2 factorial arrangement with five replicates per treatment.

Treatments consisted of two soil water availability conditions (with and without water deficit) and two CM application conditions (with and without CM). Experiment 1 was conducted using the hybrid *Urochloa* cv. Sabiá, while Experiment 2 was conducted using the hybrid *Urochloa* cv. Cayana. Each experimental unit consisted of one 8 dm³ pot containing eight plants.

2.2. Preparation of the Carbon-Based Material

Carbon dots are nanomaterials belonging to the class of carbon-based photoluminescent particles (CPP) and are generally characterized by a zero-dimensional and quasi-spherical morphology [15]. In the present study, carbon-based material (CM) was synthesized according to the method described by Vieira et al. [16], who systematically investigated the synthesis of CPP using glycerol as a precursor and characterized their photoluminescent and morphological properties.

For CM synthesis, an aqueous solution containing pure glycerol (Sigma-Aldrich, Barueri, SP, Brazil) at 90% (*w/w*) relative to water (90 g glycerol:10 g water) was prepared. Citric acid was added at 1% (*w/w*) relative to the amount of glycerol (0.9 g). The citric acid (Sigma-Aldrich, Barueri, SP, Brazil) was dissolved in water and subsequently added to the previously weighed glycerol. The synthesis was carried out in an open system using conventional heating for 2 h at 200 °C.

The resulting material was used to prepare the solution applied to the tropical forage grasses. For this purpose, an aqueous CM solution was prepared at a final concentration of 500 mg L⁻¹. The same batch of CM was used in both experiments.

2.3. Experimental Establishment

The soil used in both experiments was collected from the 0–20 cm layer of a Red Oxisol, which had previously been cultivated with maize. After collection, the soil was sieved through a 4 mm mesh to remove large particles, and a subsample of the sieved soil was used for chemical and granulometric analyses (Table 1). The remaining sieved soil was homogenized and transferred into pots. Phosphorus fertilization was then performed by applying 100 mg dm⁻³ of phosphorus in the form of monoammonium phosphate (36% P₂O₅), incorporated into the 0–2 cm surface layer. Immediately after fertilization, 20 seeds were sown per pot. Ten days after sowing, thinning was carried out to maintain eight uniform plants per pot. Subsequently, nitrogen fertilization was applied at 50 mg dm⁻³ in the form of urea.

Table 1. Chemical and particle size analysis of the Oxisol prior to initial fertilization.

pH	P *	K	Ca + Mg	Al + H	CEC	V	M	Sand	Silt	Clay
CaCl ₂			cmol _c dm ⁻³			%			%	
6.30	1.6	103	4.7	1.8	6.3	73	0	416	208	376

* Mehlich–1 method; CEC: cation exchange capacity; V: base saturation; M: aluminum saturation.

Forty days after sowing, in both experiments, a uniformity cut was performed at a residual height of 15 cm. Then, a nitrogen fertilization at 100 mg dm⁻³ in the form of urea was performed, followed by the application of the CM. The stock CM solution had a concentration of 500 mg L⁻¹. For foliar application, 50 mL of the stock solution was diluted to a final spray volume of 60 L, corresponding to 0.83 mL of stock solution L⁻¹ of spray solution. The application rate and spray volume were based on the recommendations for the commercial carbon-based nanotechnology product Arbolin Biogenesis® (Krilltech, Brasília, Brazil). The volume applied to each experimental unit was scaled considering a reference pasture density of 15 plants m⁻² [17]. Based on a spray volume of 60 L ha⁻¹, the calculated volume was 3.2 mL per pot containing eight plants; therefore, approximately 3 mL of the diluted solution was applied to each experimental unit.

In both experiments, water-stressed treatments were irrigated only once per week, bringing the soil moisture up to its maximum water-holding capacity, whereas non-stressed treatments were irrigated daily. The maximum water-holding capacity of the soil was determined using the gravimetric method [18]. The amount of water applied at each irrigation event was determined by weighing the pots. Soil moisture was not monitored between irrigation events.

2.4. Data Collection

Thirty days after the uniformity cut, plant height was measured using a graduated ruler, and the number of tillers was recorded for each experimental unit. Chlorophyll index was assessed on the middle portion of three recently fully expanded leaves using a chlorophyll meter (ClorofiLOG/CFL 1030; Falker, Porto Alegre, Brazil). Then the first harvest of the forage was performed, leaving a residual height of 15 cm. With the harvested material, the number of leaves was counted, and the whole harvested material was weighed, dried in a forced-air oven at 55 ± 5 °C for 72 h, and then weighed again for calculation of dry forage mass. Due to the morphological characteristics of the grasses utilized in this study, the harvested material was composed exclusively of leaf blades.

After the first forage harvest, approximately 3 mL of the diluted CM solution described above was reapplied to each experimental unit, together with nitrogen fertilization at 100 mg dm^{-3} in the form of urea. Thirty days later, the same procedure was repeated, and the second harvest was performed, resulting in a total of two evaluated growth cycles.

In the second harvest, the residual forage mass was harvested at ground level, weighed, and dried according to the procedure described above. Then, the content of each experimental unit (pot) was placed on sieves, and the soil was washed out using a high-pressure water machine until the roots were apparently clear of soil and dirt. Subsequently, the roots were weighed and dried as described above.

As the harvested forage consisted exclusively of leaf blades, the individual mass of each leaf was estimated by dividing the forage mass by the number of leaves. The mean number of leaves per tiller was calculated by dividing the number of leaves by the number of tillers. The phyllochron was defined as number of days required for the emergence of a new leaf. The variation in forage mass under water deficit was calculated as the difference between the mass obtained under non-deficit conditions and that obtained under deficit conditions.

2.5. Statistical Analysis

Statistical analyses were performed separately for each grass because the experiments were conducted independently. For variables evaluated in both regrowth cycles, water availability and CM application, as well as their interaction, were considered fixed effects, and the two regrowth cycles were considered repeated observations within the same experimental unit (pot). A compound symmetry covariance structure was used to account for the correlation between successive observations within each pot [19]. The following statistical model was used:

$$Y_{ijkl} = \mu + W_i + C_j + (W \times C)_{ij} + \varepsilon_{ijkl}$$

where Y_{ijkl} is the observed value in the l th regrowth cycle of the k th pot; μ is the overall mean; W_i is the fixed effect of water availability; C_j is the fixed effect of CM application; $(W \times C)_{ij}$ is the fixed interaction between water availability and CM application; and ε_{ijkl} is the residual error. The two regrowth cycles were considered repeated observations within the same pot, and a compound symmetry covariance structure was used to account for the correlation between observations from the same experimental unit. Observations from different pots were considered independent.

Root mass and residual forage mass were determined only at the end of the second regrowth cycle and were therefore analyzed without a repeated-measures structure. For these variables, the following model was used:

$$Y_{ij} = \mu + W_i + C_j + (W \times C)_{ij} + \varepsilon_{ij}$$

where the terms are defined as above. Residual normality was assessed using the Shapiro–Wilk test, and homogeneity of variances was assessed using Levene’s test. No data transformation was required. When significant effects were detected, means were compared using Tukey’s test at the 5% probability level. Statistical analyses were performed using R software (version 4.6.1, R Core Team, Vienna, Austria) [20].

3. Results

3.1. Experiment 1—Sabiá Grass

For Sabiá grass, CM application and water deficit affected several growth and productive traits, while significant $\text{CM} \times \text{WD}$ interactions were observed only for plant height and phyllochron (Table 2). CM application increased forage mass of Sabiá grass by approximately 19% regardless of water availability. It also increased leaf number, chlorophyll index,

and individual leaf mass, whereas root and residual forage mass were not affected (Table 3). Water deficit reduced forage mass by approximately 29%, accompanied by reductions of approximately 11% in tiller number and 19% in leaf number, but did not affect root or residual forage mass (Table 3).

Table 2. Summary of the analysis of variance for the effects of CM, water deficit, and their interaction on Sabiá grass.

Variables	CM	Water Deficit (WD)	CM × WD	SEM
Height	<0.001	0.0017	0.0282	0.739
Tillers	0.7950	0.0138	0.9360	1.970
Leaves	0.0498	0.0373	0.2400	4.811
Forage mass	0.0002	<0.001	0.0913	0.566
Residual forage mass	0.1636	0.2852	0.7067	1.364
Root mass	0.3720	0.1453	0.8832	4.050
Chlorophyll index	0.0472	0.0763	0.1530	1.567
Individual leaf mass	0.0443	0.1750	0.4880	0.005
Phyllochron	<0.001	0.7950	0.0020	0.302

CM: Carbon-based material; SEM: standard error of the mean.

Table 3. Productive traits and chlorophyll index of Sabiá grass subjected to CM application and water deficit.

Variable	WD−/CM−	WD−/CM+	WD+/CM−	WD+/CM+
Tillers (number pot ^{−1})	46.9 ± 2.79	46.3 ± 2.17	42.3 ± 4.26	41.3 ± 0.66
Leaves (number pot ^{−1})	126.0 ± 5.86	129.7 ± 6.75	95.2 ± 8.50	113.2 ± 5.74
Forage mass (g pot ^{−1})	20.1 ± 0.85	21.6 ± 0.71	13.0 ± 1.02	16.9 ± 0.54
Root mass (g pot ^{−1})	30.36 ± 8.00	25.95 ± 6.33	22.44 ± 5.02	16.33 ± 1.43
Residual forage mass (g pot ^{−1})	18.99 ± 2.74	21.07 ± 1.59	16.12 ± 1.91	19.68 ± 1.09
Chlorophyll index	36.59 ± 1.59	37.82 ± 1.43	34.57 ± 3.44	41.47 ± 1.81
Individual leaf mass (g)	0.160 ± 0.008	0.164 ± 0.009	0.139 ± 0.005	0.151 ± 0.005

Values are presented as means ± SEM ($n = 5$). WD−: without water deficit; WD+: with water deficit; CM−: without carbon-based material; CM+: with carbon-based material.

Under non-deficit conditions, CM application did not affect plant height, whereas under water deficit, plants treated with CM were taller than untreated plants (Table 4). Similarly, CM application reduced phyllochron under water deficit but had no effect under non-deficit conditions. Consequently, phyllochron increased under water deficit in plants without CM but remained unchanged in CM-treated plants (Table 4).

Table 4. Interaction between CM application and water deficit on plant height and phyllochron of Sabiá grass.

CM	Water Deficit		Water Deficit	
	Without	With	Without	With
	Height (cm)		Phyllochron (Days Leaf ^{−1})	
Without	40 ± 0.70 aA	31 ± 0.87 bB	9 ± 0.49 aB	12 ± 0.41 aA
With	42 ± 1.25 aA	37 ± 0.84 aB	9 ± 0.35 aA	9 ± 0.22 bA

Values are presented as means ± SEM ($n = 5$). Means followed by the same uppercase letter within rows and the same lowercase letter within columns do not differ according to Tukey's test ($p > 0.05$). CM: carbon-based material.

3.2. Experiment 2—Cayana Grass

For Cayana grass, CM application affected residual forage mass and individual leaf mass, while water deficit affected residual forage mass and chlorophyll index. A significant

CM $\times$ WD interaction was observed only for chlorophyll index (Table 5). Plant height, tiller number, leaf number, forage mass, root mass, and phyllochron were not affected by CM application or water deficit (Tables 5 and 6). Residual forage mass was approximately 14% lower with CM application and 17% lower under water deficit, whereas individual leaf mass was approximately 5% higher with CM application (Table 6).

Table 5. Summary of the analysis of variance for the effects of CM, water deficit, and their interaction on Cayana grass.

Variables	CM	Water Deficit (WD)	CM $\times$ WD	SEM
Height	0.9998	0.5200	0.2140	0.922
Tillers	0.0710	0.1830	0.2840	1.699
Leaves	0.3800	0.1730	0.9930	4.557
Forage mass	0.6520	0.6260	0.4030	0.803
Residual forage mass	0.0277	0.0031	0.8517	0.911
Root mass	0.2025	0.5910	0.7978	2.559
Chlorophyll index	0.0717	0.0040	0.0165	1.443
Individual leaf mass	0.0371	0.7150	0.3240	0.006
Phyllochron	0.3830	0.2870	0.2210	0.388

CM: carbon-based material; SEM = standard error of the mean.

Table 6. Productive traits of Cayana grass subjected to CM application and water deficit.

Variable	WD−/CM−	WD−/CM+	WD+/CM−	WD+/CM+
Height (cm)	37.3 $\pm$ 1.66	34.4 $\pm$ 0.73	35.0 $\pm$ 1.72	35.0 $\pm$ 0.74
Tillers (number pot ^{−1})	44.5 $\pm$ 2.36	43.3 $\pm$ 2.15	45.5 $\pm$ 2.89	39.7 $\pm$ 2.13
Leaves (number pot ^{−1})	123.0 $\pm$ 5.77	116.1 $\pm$ 7.61	112.7 $\pm$ 7.92	105.9 $\pm$ 3.48
Forage mass (g pot ^{−1})	18.94 $\pm$ 1.49	17.77 $\pm$ 1.11	16.60 $\pm$ 0.87	17.13 $\pm$ 0.98
Root mass (g pot ^{−1})	29.47 $\pm$ 1.43	26.54 $\pm$ 5.13	23.72 $\pm$ 2.98	22.68 $\pm$ 3.90
Residual forage mass (g pot ^{−1})	24.81 $\pm$ 1.50	21.44 $\pm$ 1.42	20.09 $\pm$ 0.52	17.21 $\pm$ 1.45
Individual leaf mass (g)	0.159 $\pm$ 0.011	0.158 $\pm$ 0.011	0.140 $\pm$ 0.007	0.154 $\pm$ 0.002
Phyllochron (days leaf ^{−1})	9.13 $\pm$ 0.34	9.55 $\pm$ 0.30	11.14 $\pm$ 0.61	10.36 $\pm$ 0.79

Values are presented as means $\pm$ SEM ($n = 5$). WD−: without water deficit; WD+: with water deficit; CM−: without carbon-based material; CM+: with carbon-based material.

Under water deficit, CM application increased the chlorophyll index, whereas no effect of CM was observed under non-deficit conditions (Figure 1). In plants without CM, the chlorophyll index remained similar under both water conditions. In contrast, in CM-treated plants, the chlorophyll index was higher under water deficit than under non-deficit conditions.

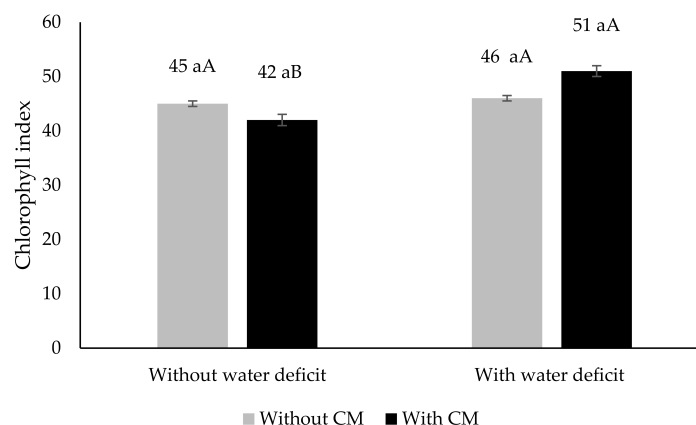


Figure 1. Chlorophyll index of Cayana grass subjected to CM application and water deficit. Means followed by the same uppercase letter within CM and the same lowercase letter within water deficit do not differ according to Tukey's test ($p > 0.05$).

4. Discussion

The two evaluated hybrid grasses showed contrasting responses to the water-deficit regime imposed in the present study. Cayana grass maintained forage production under water deficit, whereas Sabiá grass showed reductions in tillering, leaf number, and forage mass. CM application also resulted in different productive responses between the two experiments, increasing forage mass in Sabiá grass but not in Cayana grass. These contrasting patterns suggest that the effects of CM may be associated with grass genotype and with the plant response to the water-deficit conditions imposed.

For Sabiá grass, water deficit reduced tillering, number of leaves, and forage mass (Table 3). However, CM application increased forage mass by 19% regardless of water availability and mitigated the effects of water deficit on plant height and phyllochron (Table 4). These results indicate that CM supported vegetative growth in Sabiá grass. Possible mechanisms underlying these responses include enhanced light capture and conversion efficiency [21] and improved photosynthesis [6,22], as previously reported in other crops [7,8,23]. However, these physiological mechanisms were not directly evaluated in the present study.

Previous studies have proposed that carbon dots can act as auxiliary light harvesters, converting ultraviolet radiation into photosynthetically active wavelengths and potentially enhancing electron transport, CO₂ assimilation, and energy conversion in photosystem II [2]. These previously reported mechanisms may help explain the lower phyllochron observed in Sabiá grass under water deficit. However, because photosynthetic rate, gas exchange, and photosynthetic efficiency were not measured in the present study, these mechanisms cannot be confirmed by our results.

CM application increased the chlorophyll index of Sabiá grass. In maize, CM have been reported to increase light conversion efficiency and chlorophyll content [8]. CM application also increased ATPase activity by 41% and NADH production by 110%, both of which are associated with cellular energy metabolism [8]. These previously reported physiological responses may help explain the increases in plant height and forage mass observed in Sabiá grass, as CM may affect processes associated with energy metabolism and carbon assimilation without necessarily altering chlorophyll content.

In addition to their potential effects on photosynthesis and energy metabolism, carbon dots have been reported to improve plant tolerance to abiotic stress by reducing oxidative damage through the activation of antioxidant enzymes such as superoxide dismutase, peroxidase, and catalase, and by regulating osmolyte accumulation and aquaporin expression [2,24]. Recent studies in maize under water deficit provide experimental support for these responses, showing that carbon dots can reduce oxidative damage and affect processes associated with water regulation, including antioxidant activity, osmolyte accumulation, and aquaporin activity [25,26]. Such mechanisms may have contributed to the responses observed in Sabiá grass under water deficit, particularly the maintenance of leaf emergence. However, these physiological and biochemical processes were not evaluated in the present study, and their involvement therefore remains hypothetical.

Cayana grass showed greater productive stability under the water-deficit regime used in the present study. Among the productive traits, only residual forage mass was affected by water deficit (Table 6), while forage mass remained relatively stable across treatments. However, this response contrasts with previous findings for this hybrid, in which water deficit reduced forage and root mass by approximately 75 and 90%, respectively [13]. This apparent discrepancy may be related to differences in the intensity and dynamics of water restriction.

In the previous study, soil moisture was continuously maintained at 30% of maximum water-holding capacity for 47 days, whereas in the present study, plants subjected to water deficit were irrigated once a week to maximum water-holding capacity. Thus,

the present irrigation regime imposed cyclic periods of water restriction followed by rehydration, which may have allowed Cayana grass to maintain forage production under the imposed conditions. Although the irrigation protocol was the same for both grasses, soil moisture between irrigation events and evapotranspiration were not measured. Therefore, differences in water use between the grasses and, consequently, in the actual intensity of water deficit experienced by the plants cannot be excluded. CM application did not significantly affect forage mass or its response to water deficit, indicating that a mitigating effect of CM on forage production under water deficit could not be demonstrated for Cayana grass under the conditions of the present study.

In Cayana grass, water deficit did not significantly affect forage mass, although it reduced residual forage mass. CM application also reduced residual forage mass without affecting harvested forage mass. Residual biomass is relevant to plant recovery after defoliation because the remaining tissues contribute to photosynthetic activity and contain reserves that can support subsequent regrowth [27–29]. Therefore, the reduction in residual forage mass observed with CM application may have agronomic implications. However, residual forage mass was measured only at the end of the second regrowth cycle, and no subsequent regrowth was evaluated. Thus, it is not possible to determine whether the lower residual biomass affected plant recovery or subsequent forage production.

The interaction between CM and water deficit was observed only for the chlorophyll index (Figure 1), which increased when both factors were present. However, this increase did not translate into productive gains, reinforcing that the greater productive stability of Cayana grass under the water-deficit regime used in the present study may have limited the detectable benefits of CM.

Overall, the results suggest that the response to carbon-based material may depend on grass genotype and the conditions under which water deficit occurs. However, the present study was conducted under controlled greenhouse conditions using only two hybrid grasses and a single carbon-based material dose. The response to carbon-based material may also depend on application rate, as different concentrations have resulted in distinct growth and physiological responses under water deficit in other plant species [25,30].

Thus, the contrasting responses observed in Sabiá and Cayana grasses should not be generalized to other forage grasses or production conditions. Further studies should evaluate different forage genotypes and carbon-based material doses and determine whether the responses observed under controlled conditions are maintained under field conditions. Such studies will be important to establish the potential of carbon-based material as a complementary technology for tropical pasture production under water-limited conditions.

5. Conclusions

Water deficit negatively affected the growth and forage production of Sabiá grass, whereas Cayana grass maintained forage production under the water-deficit regime evaluated in this study. Carbon-based material application increased forage mass in Sabiá grass regardless of water availability and mitigated the effect of water deficit on phyllochron, contributing to the maintenance of leaf emergence under water-limited conditions. In contrast, carbon-based material did not improve forage production in Cayana grass. These contrasting responses suggest that the effects of carbon-based material may depend on grass genotype and the conditions under which water deficit occurs. Further studies evaluating different forage genotypes and carbon-based material doses under field conditions are needed to determine the consistency and agronomic relevance of these responses.

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