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Grazing Intensity Impacts on Herbage Mass, Sward Structure, Greenhouse Gas Emissions, and Animal Performance: Analysis of Brachiaria Pastureland

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Abstract: A 7 year experiment (2008–2014) evaluated cattle grazing intensity (sward height) effects on herbage mass, forage quality, and greenhouse gas emissions in continuously stocked pastures containing the tropical 'Marandu' palisade grass (*Brachiaria brizantha* (Hochst. ex A. Rich) Stapf cv. Marandu). The experiment consisted of three sward height treatments (15, 25, and 35 cm) and six replicates. There were four periods each year during the rearing phase. Significant effects were found for herbage mass, proportions of leaf and stem, crude protein, neutral detergent fiber, acid detergent fiber, lignin, animal performance, enteric methane (CH₄), and greenhouse gas (GHG) emissions from soils. When the canopy height increased from 15 to 35 cm, the herbage mass rose from 5.23 to 9.86 kg t ha⁻¹, leaf percentage decreased, and stem percentage increased. Crude protein content averaged 14.2%, and neutral detergent fiber averaged 58%. Average daily gain averaged 0.67, 0.81, and 0.90 kg ⁻¹ head⁻¹, while live weight gain ha⁻¹ was 649, 530, and 439 kg for the 15, 25, and 35 cm treatments, respectively. The weather variables explained the GHG emissions, interannual herbage mass, and structure variations.

Keywords: beef cattle; tropical pasture; Marandu grass; continuous stocking

1. Introduction

Beef cattle production is an important source of protein, minerals, and vitamins for human nutrition. However, it requires significant quantities of natural resources and may negatively impact the environment through greenhouse gas (GHG) emissions [1–3]. Beef cattle production is pasture-based

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in tropical regions, but tropical grasses usually have low crude protein (CP), and high neutral detergent fiber (NDF) and acid detergent fiber (ADF) concentrations, which lead to lower animal performance and greater enteric methane (CH₄) emissions [2,4].

The genus *Brachiaria* (syn. *Urochloa*) has been extensively cultivated in tropical regions. In Brazil, more than 100 million hectares are cultivated with *Brachiaria*, mainly 'Marandu' (*Brachiaria brizantha* (Hochst. ex A. Rich) Stapf.). *Brachiaria* species account for 60% of the Brazilian market for forage seeds and 90% of exported seeds [5]. Reasons for the success of *Brachiaria* are the adaptation and persistence of these grasses in soils with low pH associated aluminum toxicity, phosphorus (P) deficiency, and low soil organic matter [6]. Therefore, this study investigated the Marandu palisade grass cultivar in a long-term study.

According to several studies, the Marandu palisade grass sward heights used in rotational grazing are 30 cm to put the animals and 15 cm to remove the animals [7–10]. These studies revealed that nutritive value decreases above 30 cm, and regrowth declines below 15 cm. Usually, the adoption of pasture height that represents 95% of light interception results in a greater green leaf proportion, CP, lesser dead material, and neutral detergent fiber (NDF) during the growing season [10]. Pasture height also affects grazing behavior [2]. Dry matter intake is only the principal determinant of animal performance [4,5,7]. Therefore, grazing management that is optimized for forage dry matter intake (DMI) with a high nutritive value is desired. Based on this, a study was conducted using a range of sward heights to evaluate grazing intensity impacts when managing Marandu palisade grass.

Grazers affect the cycling of carbon (C) and nitrogen (N) within pastures via defoliation, excretal returns, and trampling. In pastures, animals emit carbon dioxide (CO_2) via their metabolic activity and CH_4 through enteric fermentation [11]. Nitrous oxide (N_2O) is also emitted when excreta and forage residues decompose. Greenhouse gases from the soil are affected by soil structure, particularly compaction, which affects soil moisture, oxygen (O_2) availability, and animal emissions [3,12,13]. Grazing Marandu to a 15 cm sward height implies a greater stocking rate, and hence more excretal returns and soil trampling by the animals. One of the assumptions investigated by this study was that CH_4 and N_2O emissions per area would decrease, while CO_2 emissions per area would rise with increasing sward heights.

Enteric CH_4 is produced in the rumen by methanogenic Archaea that use the hydrogen (H_2) produced during the feed fermentation process. Methanogenesis is essential for the optimal performance of the rumen because it avoids H_2 accumulation [14]. The main feed components that affect fermentation by microbiota are the fiber components (cellulose and hemicellulose), which vary when the forage sward is managed at different heights. Therefore, sward height may affect enteric CH_4 production. There have been few previous reports on GHG emissions from cattle grazing tropical pastures. For national GHG inventories in Latin America, the Intergovernmental Panel on Climate Change (IPCC) guide recognized an emission factor default of 56 kg CH_4 head $^{-1}$ year $^{-1}$ for adult beef cattle [15]. Therefore, it was hypothesized that daily CH_4 methane emissions per animal would decrease with increasing sward heights and would differ from the IPCC emission factors.

The relationships among forage production, canopy structure, nutritive value, and animal performance are complex according to long-term studies. The grazing intensity may have the greatest effect because it is the most important component of grazing management. The aims of this study were (1) to analyze the effects of different sward heights on herbage mass, plant morphological structure, herbage nutritive value, soil and animal GHG emissions, and animal performance; (2) identify relationships among variables that explain the variation in forage nutritive value and animal performance; and (3) verify which variables best explain GHG production in the soil and how these emissions are affected by season. There were two main hypotheses underlying the study. The first was that maintaining a lower sward height (15 cm) would result in a greater herbage nutritive value and animal production per area, while a sward height of 25 cm will produce the best relationship between herbage mass, animal production per area, and animal characteristics. The second was that the fiber and lignin concentrations and the proportion of stem and dead material would increase

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with pasture height. The analysis used data from a 7 year experiment on a tropical grass; Marandu palisade grass.

2. Materials and Methods

2.1. Experimental Site and Grazing Management

The data originated from a series of experiments carried out from 1 December 2007 to 31 March 2014. The experimental site was located within the Forage and Grasslands sector at São Paulo State University, "Julio de Mesquita Filho" (UNESP) (Jaboticabal, São Paulo, Brazil). The climate in this region is classified as a humid subtropical type with wet summers and dry winters. The average annual rainfall is 1424 mm, and the average air temperature is 22.3 °C. Rainfall and air temperature were measured during the experiment (Table 1). The soil is a Rhodic Ferralsol [16] derived from basalt, and the pasture was planted with Marandu palisade grass in 2001. The experimental area from 2008 to 2014 was divided into 18 experimental paddocks. Six paddocks per treatment with 0.7, 1.0 and 1.3 ha for 35, 25, and 15 cm, respectively. The reserve area was 5.0 ha in size.

D ' 1	Days with Rain	Rainfall	Temperature (°C)		
Period	(d)	(mm)	Max.	Min.	Mean
December 2005/May 2006	82	1089	31.3	17.2	23.2
December 2006/May 2007	95	1009	317	18.7	24.2
December 2007/May 2008	86	1145	30.3	14.2	22.8
December 2008/May 2009	87	1105	32.1	17.2	23.4
October 2010/May 2011	92	1455	31.1	17.7	23.1
January 2012/May 2012	47	454	31.8	18.2	25.0
December 2012/May 2013	75	1047	29.8	18.4	23.1
October 2013/April 2014	78	832	32.2	17.1	24.1

Table 1. Days (d) with rain, rainfall (mm), and average air temperatures during the experimental period.

The pasture was fertilized on a regular basis according to the recommendations from the Committee for Fertilization and Liming of São Paulo State after considering the soil chemical analyses and forage production targets [17]. In the studied area, fertilization occurred during the rainy season in the Months of November to March. The fertilizer application schedule is summarized in Table 2 based on soil chemical analyses conducted every year.

In the studied region, the forage growing season began in October and finished in April. During the dry period, the animals remained in the field and received silage and concentrates, or the stocking rates were reduced by the farms. In the period of May to September, 11% of the slaughtered animals were finished in the feedlots. In the present study, the pasture was continuously stocked with young bulls (*Bos indicus* var. Nellore), and Nellore crosses with other *B. indicus*. The animals arrived at the experimental area in October and November. In November, the pastures achieved the desired sward height. In general, the experiments commenced in December after 15 d of adaptation and were conducted for 84 or 112 days (3 or 4 periods of 28 day).

The bulls used in the experiments were 10–12 months old and weighed 180–230 kg when the evaluations began. They were identified, weighed, and randomly distributed in groups of six bulls "testers" per paddock (108 experimental animals). Some animals (40–45 put and take animals) were used to maintain the sward height using the put-and-take method [18]. In the put-and-take method, there are fixed animals "testers" and put and take animals used to adjust the stocking rate according to the herbage accumulation (mass production) to keep the management target (sward height in this study). The height/herbage mass relationship (bulk density) was used weekly to adjust the stocking rate by use of the put and take animals.

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Year	Fertilization and Lime Application	Top-Dressed Fertilization	Fertilization Schedule
2006		$200 \mathrm{kg} \mathrm{ha}^{-1} \mathrm{N}$ (urea and $160 \mathrm{kg}$ $\mathrm{ha}^{-1} \mathrm{K}_2\mathrm{O}$ (Potassium chloride)	split over two applications
2007	50 , 25 , and 50 kg ha^{-1} of NPK	$50 \mathrm{kg} \mathrm{ha}^{-1} \mathrm{N} \mathrm{(urea)}$	one application
2008	50, 25, and 50 kg ha ⁻¹ of NPK	250 kg ha ⁻¹ N (urea)	split over three applications
2009	12, 42, and 24 kg ha ⁻¹ of NPK	$250 \text{ kg ha}^{-1} \text{ N (urea)}$	split over two applications
2011	8, 28, and 16 kg ha^{-1} of NPK	$180 \text{ kg ha}^{-1} \text{ N (urea)}$	split over four applications
2012	25, 25, and 25 kg ha ⁻¹ of NPK	$90 \text{ kg ha}^{-1} \text{ N (urea)}$	split over two applications
2013	7, 20, and 14 kg ha^{-1} of NPK	160 kg ha ⁻¹ N (urea)	split over three applications
2014	$1000 \ \mathrm{kg} \ \mathrm{ha}^{-1} \ \mathrm{of} \ \mathrm{dolomitic} \ \mathrm{lime}$ and $4 \ \mathrm{and} \ 62 \ \mathrm{kg} \ \mathrm{ha}^{-1} \ \mathrm{of} \ \mathrm{N} \ \mathrm{and} \ \mathrm{P}$	$180~{\rm and}~40~{\rm kg}~{\rm ha}^{-1}~{\rm of}$ formulated N and K	split over four applications

Table 2. Description of pasture fertilization during the experimental period. N: nitrogen; P: phosphorus; K: potassium.

2.2. Experiments and Treatments

From 2008 to 2014, the treatments were three sward heights (15, 25, and 35 cm) in a completely randomized block with six replications per treatment. The factor of blocking was soil topography.

2.3. Response Variables

Herbage mass, sward canopy structure, herbage nutritive value, and animal performance were measured between 2008 and 2014; forage intake was quantified between 2007 and 2014; enteric CH_4 was measured between 2012 and 2014; and soil GHG emissions and other explanatory variables were quantified in 2013 and 2014. The source of the data was the nine dissertations or theses listed in the Supplementary Information SI.

Grazing heights were measured weekly using a graduated rule at 80 random points ("hits") per paddock and the data were used to estimate average sward height. The forage mass was estimated by clipping eight $0.25~\text{m}^2$ quadrats at average paddock sward height locations in each pasture to a residual height of 5 cm every 28 d during the measurement period. The forage samples were separated into dead material, stem + leaf sheaths, and live leaves. Then they were dried at $55 \pm 5~\text{°C}$ to constant weight to estimate dry matter (DM) ha⁻¹. The forage nutritive value, was estimated by hand-plucking method, samples every 28 d (20 samples per plot). These were then dried at $55 \pm 5~\text{°C}$ to a constant weight and ground through a 1-mm screen in a shear mill (Thomas-Wiley Laboratory Mill Model 4, H. Thomas Co, São Paulo, Brazil.) for subsequent analyses.

Forage CP, NDF, ADF, Ether Extract, and lignin concentrations were determined using the standard procedures outlined by the AOAC (1990). Indigestible NDF was quantified by incubating hand-plucking and feces samples dried at 55 ± 5 °C to a constant weight and ground through a 2 mm for 244 in the rumen. The following equation was used to determine the apparent digestibility of DM: digestibility (%) = $100 - (100 \times (i\text{NDF in hand-plucking sample/iNDF in feces}) \times (nutrient concentration in feces/nutrient concentration in feedstuff) Enteric CH₄ was measured using a tracer gas method (sulfur hexafluoride—SF₆) in 2012, 2013, and 2014. The complete methodologies are described in Barbero et al. (2015) [19].$

Animal performance variables were calculated as follows [18]: Body weight gain—BW gain (kg) = (final BW – initial BW);

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Average daily gain—ADG (kg animal⁻¹ day⁻¹) = (BW gain (kg)/days);

Gain per area—GPH = $(ADG \times number of animals per days \times experimental period (days))/area (ha), where the number of animal days (animal day⁻¹ ha⁻¹) was calculated by dividing animal stock by the mean weight of "testers". Animal stock was determined by the sum of weights of all animals present in each paddock divided by the area of the paddock (kg BW ha⁻¹);$

Stocking rate in animal unit (AU = 450 kg BW)/ha = $(\sum BWmean/450)$ /area (ha);

Greenhouse gases from the soil were quantified using the static closed chamber method described by Cardoso et al. (2017) [12]. The explanatory variables for soil moisture, ammonium, and nitrate were quantified at the 0–20 cm soil depth. Soil samples of the 0–10 cm layer were sampled at each air collecting event to quantify mineral N, gravimetric water content (by drying soil at $105\,^{\circ}$ C), and percent water filled pore space (WFPS). Soil bulk density in the 0–10 cm layer was determined using a cylinder 50 mm in diameter and 50 mm in height. WFPS was calculated from the gravimetric water content and bulk density using a particle density of 2.65 g cm⁻³. For mineral N analysis, extraction with 2 M KCl was done on field moist samples with correction for water content. Ammonium-N was determined using the Berthelot reaction with spectrometry at 647 nm. Nitrate-N quantification was carried out by ultraviolet absorption spectrometry at 220 [12]. Air temperatures, moisture, precipitation, and solar radiation were obtained from a meteorological station located 500 m away from the study area.

2.4. Statistical Analysis

Data were analyzed as repeated measures using the LME procedure in R (package NLME, R Core Team, https://svn.r-project.org/R-packages/trunk/nlme/). The model included sward height, years, and sward height versus year interactions as fixed effects, and paddock or animal as the random effect. Non-structured covariance was used for the repeated-measures analyses because it had the lowest Akaike Information Criterion. Orthogonal polynomial contrasts were performed when a significant effect was found.

Principal component analysis (PCA) was obtained from a correlation matrix using the R package "FactoMineR" (http://factominer.free.fr/). The categories analyzed were: (1). Forage mass and structure (Dry matter mass, leaf mass, stem mass, dead material mass, % of leaf, % of stem, % of dead material, and leaf/stem ratio), and (2) Nutritive value (Organic Matter%, CP%, NDF%, and lignin% of DM),

In this study, canonical correlation analysis (CCA) was used to correlate monthly observations of climate independent variables (air temperature, air moisture, rainfall and solar radiation) with dependent variables: herbage mass and structure (Dry matter mass, leaf mass, stem mass, dead material mass, % of leaf, % of stem, % of dead material, and leaf/stem ratio). And, for the GHG from soil analysis, the climate variables (mean, minimal, and maximal air temperature; mean, minimal, and maximal air moisture; rainfall; and solar radiation) were the independent variables and CO_2 , N_2O , and CH_4 fluxes were the dependent variables. Daily data were used. The best multiple regression model was chosen using R^2 and the significance of the model was calculated by the "FactoMinerR" package in R (R Core Team, 2016).

3. Results

3.1. Forage Mass and Structure

Pasture height affected herbage mass (p < 0.0001), leaf mass (p < 0.0001), stem mass (p < 0.0001), dead material mass (p < 0.0001), % of leaves in the herbage (p < 0.006), and % of stems (p < 0.0158), but not the percentage of dead material (p = 0.29). Herbage mass varied from 3460 DM ha⁻¹ in 2009 in the 15 cm treatment to 11,000 kg DM ha⁻¹ in 2013 in the 35 cm treatment. During the seven years of evaluation, herbage mass averaged 5230, 7810, and 9860 kg DM ha⁻¹ for the 15, 25, and 35 cm treatments, respectively. Herbage mass increased linearly with sward height.

The lowest leaf mass was recorded in 2012 (1330 kg DM ha^{-1}) and the highest in 2013 (4240 kg DM ha^{-1}) (Table 3). The leaf mass range was 1760, 2460, and 3010 kg DM ha^{-1} for the 15,

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25, and 35 cm sward heights, respectively. The average stem mass at 35 cm height (3560 kg DM ha^{-1}) was more than double the stem mass recorded at 15 cm (1490 kg DM ha^{-1}) and was greater than at 25 cm (2410 kg DM ha^{-1}). The greatest mass of dead material was found in 2008 and the lowest in 2013. The dead material mass averaged 1940, 2830, and 3220 kg DM ha^{-1} , respectively, for the 15, 25, and 35 cm sward heights (Table 3). Leaf, stem, and dead material mass increased linearly with sward height.

The forage morphological composition results showed that the average leaf proportion decreased linearly from 40% to 35.7% as sward height increased (p = 0.006); the proportion due to stems increased linearly (p = 0.0158) from 42.6% to 45.2%; and the % of dead material decreased linearly, with averages of 37.2, 35.5, and 32.6% for the 15, 25, and 35 cm sward heights, respectively, (Table 3).

3.2. Nutritive Value

The nutritive value results showed that sward height affected the organic matter (OM), NDF, and ADF values (p < 0.01). The effect of grazing intensity on CP and digestibility had p-values of 0.06 and 0.0548, respectively. Table 4 shows that OM was similar across all treatments (approximately 91%). Crude protein varied greatly among years and decreased linearly with sward height when all the year data were analyzed together. The highest CP occurred in 2011 (16.3%) in the 15 cm treatment, whereas the lowest CP content was 11.2%, which occurred in 2014. The CP content averaged 14.7, 14.0, and 14.0% for the 15, 25, and 35 cm sward heights, respectively.

Both the NDF and ADF fractions increased linearly with sward height. The highest NDF concentration was measured in 2009 for all treatments and the lowest in 2011. The average NDF values during the 7 years of evaluation were 57.8, 59.1, and 59.6% for the 15, 25, and 35 cm treatments, respectively. The ADF value for the 35 cm treatment (32.0%) was significantly higher than 15 and 25 cm sward heights, which both averaged 30.4%. Lignin content averaged 3.7, 3.8, and 4.2% for the 15, 25, and 35 cm treatments, respectively (Table 4). The results of principal component analysis identified that temperature and rainfall explained more than 80% of the variation in Marandu palisadegrass chemical composition (temperature explained 45% and rainfall 41%). The canonical correlation analysis between the weather variables and nutritive value showed that rainfall and temperature were the most correlated variables. The coefficients were 0.23 and 0.49 for rainfall and temperature, respectively. The variation in the NDF content was explained by the variation in the temperature. The coefficient were 0.94 between NDF and temperature.

3.3. Animal Performance

Sward height affected daily weight gain (p = 0.027), which increased linearly by 0.668, 0.892, and 0.902 kg Body weight (BW) day⁻¹ on average for the 15, 25, and 25 cm treatments, respectively (Table 5). The highest individual gain exceeded 1 kg BW day⁻¹ and was observed in 2013, while the lowest was recorded in 2008. The highest gain per area was recorded in 2011 and 2013 at the highest grazing intensity (15 cm sward height), and the lowest was observed in 2008 in the 35 cm treatment (196 kg BW ha⁻¹). Gain per area averaged 649, 530, and 439 kg BW ha⁻¹ and was affected by sward height (p = 0.004). This gain was recorded during the rainy season (from December to April). Stocking rate was significantly affected by sward height (p < 0.0001) and decreased linearly in response to pasture height. The stocking rate averaged 6.13, 4.73, and 4.09 animal units ha⁻¹ (1 animal unit equals 450 kg BW) for the 15, 25, and 35 cm treatments, respectively.

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Table 3. Herbage mass and structural characteristics of Marandu grass managed at different sward heights ¹.

		Sward Height (cm)				<i>p-</i> Value		
Variable	Year	15	25	35	Sward	Year	S*Y	
	2008	7.22	8.86	10.03				
Herbage mass	2009	3.46	5.08	7.45				
	2011	4.95	9.24	10.22				
	2012	4.37	7.59	9.61	<i>p</i> < 0.001	p < 0.001	p = 0.298	
$(t DM ha^{-1})$	2013	5.99	7.74	11.02		p 10.001	p 0.2>0	
	2014	5.36	8.37	10.83				
	Mean	5.23	7.81	9.86				
	2008		1.79	2.26				
	2008	1.64 1.45	2.39	2.26				
	2009	1.43	2.39	2.92				
Leaf mass	2011	1.43	2.47	2.92	n < 0.001	<i>p</i> < 0.001	n = 0.42F	
$(t DM ha^{-1})$					<i>p</i> < 0.001	p < 0.001	p = 0.435	
	2013	2.59	3.16	4.24				
	2014	2.10	2.80	3.43				
	Mean	1.76	2.46	3.01				
	2008	1.84	2.71	3.59				
	2009	1.10	1.70	2.80				
Stem mass	2011	1.44	3.46	3.65				
$(t DM ha^{-1})$	2012	1.24	2.30	2.95	p < 0.001	p < 0.001	p = 0.467	
(t Divi iia ")	2013	2.37	3.33	5.26				
	2014	1.49	2.41	3.60				
	Mean	1.58	2.65	3.64				
	2008	3.74	4.36	4.18				
	2009	1.24	1.75	2.26				
	2011	2.08	3.31	3.65				
Dead material mass	2012	1.80	3.14	3.85	p < 0.001	p < 0.001	p = 0.657	
$(t DM ha^{-1})$	2013	1.03	1.24	1.52	,	,	,	
	2014	1.76	3.17	3.84				
	Mean	1.94	2.83	3.22				
	2008	47.0	40.1					
	2008	49.8	41.5	38.8 44.5				
	2009	30.4	28.3	29.5				
0/ 16	2011	30.4	30.3	30.7	p = 0.006	<i>p</i> < 0.001	p = 0.499	
% leaf	2012	43.4	40.9	38.5	p = 0.000	$\rho < 0.001$	p = 0.49	
	2013	43.4 39.0	34.1	32.4				
	Mean	40.0	35.9	35.7				
	2008	53.0	60.0	61.0				
	2009	48.8	47.4	43.5				
0/ -1	2011	58.5	58.5 20.2	55.5 30.7	n < 0.016	n < 0.001	p = 0.626	
% stem	2012	28.3	30.3	30.7	p < 0.016	<i>p</i> < 0.001	p = 0.626	
	2013	39.5	43.1	47.8				
	2014 Mean	27.7 42.6	28.5 44.6	32.5 45.2				
	2008	50.8	48.7	41.1				
	2009	38.9	35.1	30.1				
	2011	41.5	34.2	35.4	0.50	0.001		
% dead material	2012	41.3	41.4	39.8	p < 0.29	p = 0.081	p = 0.822	
	2013	17.2	16.0	13.8				
	2014	33.3	37.4	35.1				
	Mean	37.2	35.5	32.6				

¹ Means of four evaluations between December and March of each year (n = 24). S = sward and Y = year. S*Y (sward height versus year interactions).

Table 4. Nutritive values for Marandu grass (hand plugged sample) managed at different sward heights ¹.

Sward Height (cm)								
Variable	Year	15	25	35	Sward	Year	S*Y	
	2008	90.0	90.6	90.8				
	2009	90.1	90.8	90.7				
	2011	91.4	90.7	90.9				
Organic matter (g kg ⁻¹ DM)	2012	91.5	91.6	91.7	p = 0.0014	p < 0.001	p = 0.839	
	2013	91.0	92.5	92.4				
	2014	91.3	91.3	91.4				
	Mean	90.9	91.3	91.3				
	2008	15.8	15.6	14.6				
	2009	14.7	14.5	13.6				
	2011	16.3	15.3	14.7				
Crude protein (g kg ⁻¹ DM)	2012	13.5	13.4	12.6	p = 0.06	p < 0.001	p = 0.093	
	2013	15.1	13.9	13.7				
	2014	12.7	11.2	14.7				
	Mean	14.7	14.0	14.0				
	2008	57.2	60.4	59.9				
	2009	60.6	61.1	63.0				
Neutral detergent fiber	2011	54.7	54.6	55.6				
$(g kg^{-1} DM)$	2012	57.6	57.5	58.9	p = 0.0043	p < 0.001	p = 0.753	
(g kg Divi)	2013	57.7	60.0	59.9				
	2014	59.1	61.2	60.0				
	Mean	57.8	59.1	59.6				
	2008	33.7	34.1	39.9				
	2009	28.7	28.8	30.1				
Acid detergent fiber	2011	28.5	28.4	35.6				
$(g kg^{-1} DM)$	2012	26.0	26.0	26.2	p = 0.033	p = 0.099	p = 0.006	
(g kg Divi)	2013	28.9	29.3	29.7				
	2014	34.4	34.0	34.1				
	Mean	30.4	30.4	32.0				
	2008	4.6	3.9	5.7				
	2009	4.6	4.7	5.0				
	2011	2.8	3.5	3.6				
Lignin (g kg ⁻¹ DM)	2012	3.3	3.3	3.2	p = 0.78	p < 0.01	p = 0.31	
	2013	-	-	-				
	2014	3.2	3.4	3.7				
	Mean	3.7	3.8	4.2				
	2008	75.8	78.7	77.0				
	2009	57.3	54.7	60.2				
Digestibility of dry matter	2011	59.5	56.0	68.6				
(% DM)	2012	63.8	64.0	69.0	p = 0.055	p < 0.001	p = 0.805	
(/o DIVI)	2013	72.8	70.4	68.5	,	,		
	2014	71.3	70.5	66.6				
	Mean	65.4	64.6	68.2				

 $^{^{1}}$. Means of four evaluations between December and March of each year. (n = 24). S = sward and Y = year. S*Y = sward height and year interaction.

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ble 5. Animal performance (gain per animal and area) in Marandu grass pastures managed	at
fferent sward heights ¹ .	

Treatment (cm)								
Variable	Year	15	25	35	Sward	Year	S*Y	
	2008	0.32	0.63	0.76				
	2009	0.61	0.73	0.78				
	2011	0.69	0.87	0.95				
Average daily gain (kg day ⁻¹)	2012	0.40	0.60	0.80	p = 0.027	p < 0.001	p = 0.644	
	2013	1.08	1.15	1.20				
	2014	0.91	0.89	0.91				
	Mean	0.67	0.81	0.90				
	2008	506	278	196				
	2009	744	672	576				
	2011	778	715	602				
Gain per area ($kg ha^{-1}$)	2012	492	528	468	p < 0.001	p = 0.544	p = 0.739	
1	2013	778	578	470				
	2014	597	410	322				
	Mean	649	530	439				
	2008	5.49	3.92	4.63				
Stocking rate (AU ha ⁻¹) ¹ Animal unit (AU) equals	2009	5.01	4.96	5.86				
	2011	5.33	4.32	3.20				
	2012	6.64	4.51	3.42	p < 0.001	p < 0.001	p = 0.476	
450 kg of body weight	2013	8.20	5.81	4.42				
5 , 3	2014	7.01	4.93	3.80				
	Mean	6.13	4.73	4.09				

¹ Four evaluations between December and March of each year. (n = 142). S = sward and Y = year. S*Y = sward height and year interaction.

3.4. Enteric Methane

Daily CH₄ emissions per animal were affected by sward height (p = 0.002) and year (p < 0.0001), but there was no interaction sward height and year. They increased linearly and per kg of BW gain (p = 0.012) e varied within year (p = 0.0003). In 2013, CH₄ emissions decreased with increasing average daily gain, whereas, an inverse effect was observed in 2014. A summary of the enteric CH₄ data is shown in Table 6. Enteric CH₄ averaged 117, 126, and 139 g animal⁻¹ day⁻¹ for the 15, 25, and 35 cm treatments, respectively, and the emissions per gain (g kg⁻¹ BW gain) followed a similar pattern to the daily emissions.

3.5. Greenhouse Gases

Soil GHGs were measured from 2012 to 2014 in the long-term study and the results showed that grazing intensity affected GHG emissions. Nitrous oxide decreased, CO₂ increased, and CH₄ showed a curvilinear response to increased sward height. Only the highest grazing intensity (15 cm sward height) produced net N₂O emissions. Furthermore, N₂O consumption increased with sward height (Table 7). The lowest CH₄ emissions were recorded in the 25 cm treatment, and the results showed that sward height caused a CO₂ emissions increase from 44.2 to 80 Mg CO₂ ha⁻¹, which was the largest sward height effect within the analyzed gases ($r^2 = 0.95$).

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Sward Height (cm)									
Variable	Year	15	25	35	Sward	Year	S*Y		
	2012	107	179	184					
Daily CH ₄ emissions (g animal ⁻¹ day ⁻¹)	2013	129	132	123	p = 0.002 $p <$	p < 0.0001	p = 0.193		
	2014	106	119	155		p < 0.0001			
	Mean	114	141	156					
	2012	117	191	174					
Emissions per gain $(g kg^{-1} BW gain)$	2013	119	115	101	p = 0.012 $p = 0.0003$	v = 0.459			
	2014	115	136	177	p = 0.012	p = 0.0003	p = 0.439		
	Mean	114	149	154					

Table 6. Enteric CH₄ emissions from Nellore bulls fed on Marandu grass managed at different sward heights ¹.

Table 7. Cumulative N_2O (kg N_2O -N ha^{-1}), CH_4 (kg CH_4 -C ha^{-1}), and CO_2 (Mg CO_2 ha^{-1}) emissions for Marandu grass managed at different sward heights ¹.

	Sward Height (cm) ²	<i>p</i> -Value			
	15	25	35		Effect
N_2O	$0.73 (0.69)^2$	-1.11 (1.10)	-2.64(1.05)	p < 0.01	Linear
CH_4	0.21 (0.06)	0.11 (0.05)	0.85 (0.50)	<i>p</i> < 0.01	ns
CO_2	44.2 (3.1)	59.4 (3.9)	80.0 (6)	p < 0.01	Linear

¹ 2 years of evaluation (n = 6). ² Within parentheses: standard error of the mean (\pm).

The principal components analysis for GHGs emissions and explanatory variables identified that there was a clear positive correlation between soil moisture, and CO₂ and N₂O emissions. Soil ammonium and nitrate were also correlated with these gases. However, soil CH₄ was not correlated with the variables (Figure 1). The CCA was performed to identify linear correlations between the grouping identified in the PCA analysis. The CCA showed the correlation between N_2O and CO₂ of 0.38 and with moisture of 0.48. The correlation with ammonium and nitrate was negative -0.53 and -0.49, respectively.

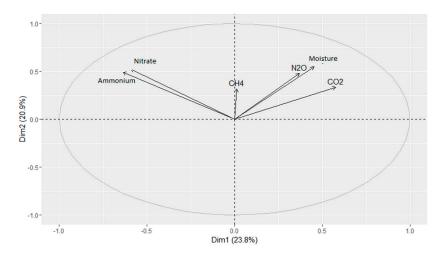


Figure 1. Principal Correlation Analysis between greenhouse gases and the explanatory variables for Marandu grass pastureland managed at different sward heights during the period 2012-2014. Dim1 (principal component 1) and Dim2 (principal component 1).

¹ One evaluation per year. (n = 18). BW—body weight, S—sward height and Y—year. S*Y = sward height and year interaction.

4. Discussion

4.1. Sward Structure

This study aimed to show how sward structure affects forage quality. Despite the difference in herbage mass over the years, the % proportions of leaves and stems had consistent relationships with sward height (Figure 2). The sward structure variables are conditioned by sward height and environmental variables (temperature, nitrogen fertilization, and light) [2,19–22]. The highest percentage proportion due to stems was recorded in the 35 cm treatment and was the result of stem elongation (Table 2).

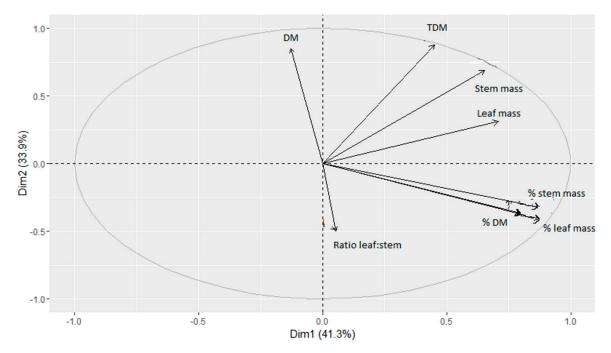


Figure 2. Principal component analysis between forage mass and the sward structure variables between 2008 and 2014. Dim 1 (principal component 1), Dim2 (principal component 2), TDM—herbage mass, DM—dead material, dead material % (% DM).

A minimal herbage mass of 2000 kg DM ha⁻¹ is required for animal selection and is the recommended amount to sustain animal gains [7]. The results from this study showed that the herbage mass varied from 5230 to 11,120 kg DM ha⁻¹, which resulted in bulk densities of 34.8, 31.2, and 28.1 g DM m⁻³ for the 15, 25, and 35 cm sward height treatments. A previous study found no differences in herbage mass among the sward heights, which averaged 4000 kg DM ha⁻¹ [19–23]. However, variations in soil fertility, rainfall, temperature, and solar radiation may explain the lower results recorded in this study compared to the previous study [23]. According to several studies, the potential production value for Marandu grass is about 11,000 kg DM ha⁻¹ year⁻¹, which was also observed in 2013 by this study (Table 2) [14]. During the 2013 growing season, the measured herbage accumulation was 58 kg DM day⁻¹ [19]. Therefore, based on 6 months growing season, approximately 10,000 kg of DM ha⁻¹ is produced based on this herbage accumulation rate.

Grazing management aims to improve sward structure (physical characteristics, tissue distribution, anatomy, and leaf mass) and is an effective strategy that can be used to improve forage quality because changes in the structure variables can promote large differences in voluntary intake, which is the main factor that determines animal performance [24,25].

4.2. Nutritive Value

The observed mean CP and NDF contents found in this study varied from 14.0% to 14.7% and from 57.8% to 59.6%, respectively (Table 3). These values differed from several previous studies that used Marandu grass [10,26,27], but were in line with a study from Vietnam [28]. In the Brazilian studies, CP varied from 10% to 12% during the rainy season (November to April), and from 6% to 8% during the dry season, whereas NDF was always above 60%.

The NDF content increased with stem proportion in the sward and as the forage matured, both of which are dependent on the environment [2,19,20]. A Pearson's correlation between NDF content and stem proportion generated a coefficient of 0.85. The increase in NDF with sward height observed in this study was probably due to variations in grass structure. Changes in plant structure alter the sward interception of photosynthetically active radiation (PAR) when the nitrogen fertilization rate is the same among treatments. The critical leaf area index concept states that when the grass sward intercepts 95% of PAR, it approaches its maximum growth rate without the plant shading itself [29]. Intercepts higher than 95% of PAR increase senescence and the stem growth rate, but decrease the grass nutritive value (decrease in CP and increase in fiber content) and leaf production [29].

The discriminatory power of variables within each principal component is measured by multiple correlations [30]. The results of principal component analysis identified that temperature and rainfall explained more than 80% of the variation in Marandu grass chemical composition (temperature explained 45% and rainfall 41%), which is in line with the findings for temperate grasses [31,32]. The simultaneous occurrence of high temperatures and drought can limit forage growth and change tissue chemical composition [32,33]. When analyzed separately, drought and temperature have been shown to alter the chemical composition of forage [33]. Elevated temperature and drought (inadequate soil moisture) change plant chemical composition by altering the concentrations of non-fibrous and fiber carbohydrates [34].

4.3. Animal Performance

In Brazil, Nellore calves are weaned at 7 months, and have an average BW of $210 \, \mathrm{kg}$ [35]. This study used animals that were bought after weaning with initial body weights ranging from $180 \, \mathrm{to} \, 230 \, \mathrm{kg}$. The effect of initial body weight on average daily gain (ADG) was attributed to the differences in the genetic group, nutrition during pregnancy, and the cow-calf phase.

Cow nutrition during pregnancy can affect the development of the progeny. Previous studies have shown that progeny from cows fed on improved pastures have higher ADGs in the finishing phase, thicker fat layers, and greater meat tenderness [36]. The development of calves from cows that were well nourished has been compared with those subjected to nutritional restrictions. The results showed that deficient nutrition can reduce the birth weight by up to 35% [37]. Furthermore, animals with lower birth weights have limited compensatory growth capabilities. In addition, when animals were slaughtered at the same age (30 months), the group with a higher birth weight showed significantly improved performances for carcass and meat cuts.

Dry matter intake (DMI) is the main variable regulating animal performance [38,39]. The DMI is controlled by numerous factors that interact with each other, such as animal characteristics, the environment, forage allowance, nutritive value, and management [40]. In this study, animal performance varied among years and sward heights.

Higher or lower ADGs and gain per hectare (GPH) values were not observed in the years with higher or lower accumulated precipitation, or the year where the grass had the highest CP (Table 1, Table 3, and Table 5), which suggests that factors inherent to animals, such as body weight, genetic factors, and feed conversion, are mainly responsible for annual variations.

Overall ADG increased with pasture height. It has been shown that grazing selectivity changes with grazing intensity [41]. In taller swards, animals can select more leaves that have higher nutritive value, which leads to a greater ADG. Over the years, pastures with taller sward heights had greater live leaf mass per hectare and total mass per hectare values. Herbage live leaf content is a key predictor of

beef cattle production because live leaves have the greatest nutritive value [10,42]. Herbage mass in this study was always greater than $5000 \text{ kg DM}^{-1} \text{ ha}^{-1}$, which meant that it was consistently higher than the 2000 kg recommended level, below which there is a limit to forage intake in *Brachiaria* pastures [7].

High intakes and animal performances are achieved when high herbage masses and allowances are provided and forage nutritive value is high. Sward height affects the number of meals per animal per day when it is less than 15 cm, while the time per meal decreased with an increase in sward height [37]. Previous studies have also shown that a larger forage allowance increases the number of meals and reduces the time per meal [20,43,44]. The improved animal performance observed in this long-term study (Table 3) suggested that both nutritional and non-nutritional factors limited forage intake [39].

This study evaluated the performance of Nellore bulls in the rearing phase during the Marandu grass growing season (December to April). They received additional mineral salts and an average supplement of 150 kg N. In Central Brazil, the ADG value without supplements is only 0.3 kg animal⁻¹ day⁻¹ [45], which is much lower than the results recorded in this study (approximately 0.8 kg animal⁻¹ day⁻¹) (Table 3). Therefore, animal performance can be increased in tropical regions by controlling sward height. The results obtained over the 7 yrs of evaluation can be attributed to (1) greater forage mass (>2 times greater than that found to limit intake), (2) a higher proportion of live leaves in the herbage mass (>50%), (3) a grazing efficiency of 50% obtained using the put-and-take method, (4) high digestibility (>60%), (5) a high CP level for tropical grasses (>140 g kg⁻¹), (6) an NDF concentration of less than 60%, and (7) an adequate protein/energy ratio (>150 g CP kg⁻¹ DOM).

Improved grassland management regimes, where the aim is sustainable intensification, need to be based on an understanding of the seasonal variations in productivity, stocking rate, sward structure, soil fertility, and animal behavior. The focus should not only be on carcass production per animal or area. Furthermore, livestock intensification needs to reduce the environmental footprint produced by cattle farming and generate ecosystem services, such as improved soil quality, reduced erosion, and GHG emissions.

4.4. Greenhouse Gases

Enteric CH₄ is affected by the chemical composition of forage. The CH₄ amount produced by animals fed on forages rich in structural carbohydrates is greater than the amount produced by those fed diets with higher levels of non-structural carbohydrates [11,46,47]. The data from this study showed that in the first and third years, the results were in line with previous studies because they showed that NDF content and enteric CH₄ production were greater in the 35 cm pasture treatment than in the other treatments (Table 6). However, the results also showed a higher variation in the treatment responses over the evaluation years. Other variables, such as animal genetics, may explain the differences between years.

The variation in CH₄ emissions from 106 to 177 g CH₄ animal⁻¹ day⁻¹ among sward heights and years can be explained by dry matter intake variation (Table 6). Similar behavior was reported in previous studies where a large variation was found between daily CH₄ emissions. This variation was attributed to forage selection by the animals and, consequently, to variations in daily dry matter intake [48,49]. In the 25 cm height treatment, the NDF concentration was lower, and digestibility and the leaf appearance rate were higher, which may explain the lower enteric CH₄ production (Table 7). Harvesting forage at an earlier grass maturity stage has often been proposed as a strategy to decrease enteric CH₄ production [50].

The IPCC guidelines for national inventories default emission factor for enteric CH $_4$ from beef cattle with the same range of body weights studied here are 149 g CH $_4$ animal $^{-1}$ day $^{-1}$ [51]. In this study, daily CH $_4$ emissions ranged from 117 to 139 g animal $^{-1}$ day $^{-1}$ (Table 6). The enteric CH $_4$ measured in this study was lower than the average IPCC emissions factor for tropical pasture (149 g animal $^{-1}$ day $^{-1}$) but within the range of variability around the mean given in the IPCC guide. The results produced by this study appear to be the first reports of enteric CH $_4$ emissions for cattle that have only consumed

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Brachiaria grass. The amount of CH₄ emitted by tropical production systems requires measurements from long-term experiments across a specified region.

Grazing intensity effects CO_2 and N_2O emissions [12]. Both GHG emissions declined in this study as the grazing intensity decreased. However, these effects varied according to the season, and there was no overall effect due to grazing intensity. The correlation between soil variables and GHG emissions was analyzed, and the results showed that there was a positive correlation between soil moisture and CO_2 and N_2O , whereas soil ammonium and nitrate were not correlated with GHG emissions (Figure 1).

Soil moisture supports biological activity [52,53] and can control CO_2 and N_2O emissions. This control is probably due to changes in root biomass, microbial decomposition, and variations in substrate supply depending on soil moisture [54]. Indeed, GHG emissions in tropical grasslands are extremely affected by seasonal variations in the rainfall regime [13,53,55]. Soil moisture also controls O_2 diffusivity and, consequently, aerobic conditions.

Nitrous oxide is produced during nitrification and denitrification reactions in the soil [56]. The results produced by this study were in line with several previous studies that reported a positive correlation between soil moisture and N_2O [13,57,58]. These studies attributed this effect to variations in N uptake by plants and microorganism populations, N transformations (mineralization, nitrification) in the soil, and, principally, increases in denitrification activity induced by reduced O_2 diffusion into the soil.

The results showed that soil NH_4^+ and NO_3^+ were not correlated with N_2O and CO_2 emissions (Figure 1). However, mineral nitrogen concentration (NH_4^+ and NO_3^-) has a controlling effect over N_2O emissions from soils [59]. The reasons for the absence of a correlation between N_2O production and mineral N in this study are not clear, but presumably, N was not a limiting factor during the experimental period, which suggests that it was soil moisture that drove the fluxes.

Methane fluxes from soil are driven by initial C and N concentrations, soil moisture, temperature, dissolved N and C, and mineral N concentration [53,60]. The results did not show a clear correlation between CH_4 fluxes and any soil variable (Figure 1). The CH_4 emissions were probably driven by different variables depending on the season and forage management activities. It has been reported that *Brachiaria* pastures are not a source of CH_4 . These previous studies suggested that the well-drained conditions of this soil favored CH_4 oxidation, which consequently offset the CH_4 production [12].

4.5. Practical Implications for Pasture Management

From a production perspective, the results were consistent with the hypothesis that increasing sward height leads to a diminution in the sward leaf and crude protein content. However, due to the higher forage allowance, there is a greater possibility of animal diet selection, resulting in higher individual performance. The feed value of Marandu grass and animal performance was relatively high in the current study.

The results also showed that the lowest sward height produced the highest N_2O emissions, which is the most potent GHG. Irrespective of pasture height, enteric CH_4 production was lower than the IPCC emissions factor for beef cattle in Latin America by approximately 30%. However, this is still within the range considered by the IPCC. The CH_4 conversion factor (Ym) factor, which represents the annual gross energy intake lost as CH_4 , was similar to that observed in high-quality forage for all sward heights. According to this IPCC guideline, high-quality forage has a Ym lower than 6% [15]. In this study, the reported Ym was approximately 5% [19].

The results showed that individual animal performance increased from 668 to 901 g day⁻¹, and gain per area decreased from 649 to 439 kg ha⁻¹ during the rearing phase. The GHG emissions during the rearing phase were calculated based on the data for enteric CH₄ reported here and GHG emissions for soil reported by Cardoso et al. (2017) [12]. The GHG data were converted into equivalent carbon dioxide (CO₂eq) using a global warming potential of 28 for CH₄ and 265 for N₂O. The analysis showed that the carbon footprint was 3.28, 3.54, and 4.08 kg CO₂eq kg⁻¹ BW gain during the rearing phase for each sward treatment. A sward height of 25 cm for Marandu grass clearly produced the best

forage quality and environmental performance according to the individual and per unit area gain in live weight results. However, there are some important aspects to consider before this management strategy is recommended. For example, both the soil type and water availability in this study were suitable for grazing. Furthermore, the put-and-take method was used to control the weekly stocking rate, which was based on the forage accumulation rate. In addition, nitrogen fertilizer was applied three or four times during the Marandu grass growing season. Despite this, the adoption of this management strategy by farms can result in high animal performances. Our results suggest that after seven years of grazing management that a fixed pasture height that corresponds to the 95% light interception during the growing season, which results in greater CP concentration and leaf proportion for Marandu palisade grass. Therefore, obtaining higher animal performance.

5. Conclusions

The principal component analysis revealed that weather-related variables had a strong effect on the structure and nutritive value of the sward. The canonical correlation analysis showed that rainfall was the main variable explaining differences in structure and nutritive value. Greenhouse gas emissions from the soil and animals were controlled by climatic variables and, to a lesser extent, by grazing intensity.

Sward height had clear effects on sward structure, nutritive value, and animal performance. Based on the data of nutritive value, animal production per area, and greenhouse gas emissions, the sward height of 25 cm is the most visible way of obtaining greater animal performance and sustainability per area using Marandu palisade grass and continuous stocking.

Further studies should quantify the interactions between N fertilization and sward height as well as on the GHG emissions from fertilized grazed grasslands. These data could form the basis for further research evaluating the potential of managing the sward height of Marandu palisade grass to achieve sustainable intensification in forage-based beef cattle production.

Supplementary Materials: The following are available online at http://www.mdpi.com/2073-4395/10/11/1750/s1. Supplementary information SI: Source of data

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