

## ORIGINAL ARTICLE

# Bahiagrass (*Paspalum notatum* CV. Inia Sepé) responses to defoliation intensities

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## Abstract

Defoliation management is determinant of forage production and plant persistence. The objective of this study was to identify grazing suitability and stability of herbage production of INIA Sepé subjected to four intensities of defoliation (represented for sward canopies of 3, 10, 17 and 24 cm maintained constant through weekly cuts) and two rates of nitrogen fertilization (100 and 300 kg ha<sup>-1</sup>). The response variables studied were: botanical/morphological composition of the herbage mass, herbage accumulation, nutritive value, tiller population density (TPD), tiller weight (TW), leaf area index (LAI) and nitrogen nutrition index (NNI) and calculations performed relative to the tiller size × density compensation mechanism (SDC). Taller swards had smaller population of large tillers, with greater individual leaf area, showing a tiller × size density compensation response. Calculations indicated that the regression coefficient for the linear regression between TW and TPD in a log scale was close to −2.5, in agreement with the −5/2 self-thinning rule, with variable LAI and R (ratio between tiller leaf area and volume). The exception was during year 1 for the 100 kg N ha<sup>-1</sup>, when the regression coefficient was close to −1.5. During year 2 only one replication of the 3 cm and 100 kg N ha<sup>-1</sup> did not follow the SDC line. LAI and herbage accumulation were positively correlated with the distance between points of log(TW) × log(TPD) and an hypothetical −3/2 SDC line ( $p < .0001$ ). *Paspalum notatum* cv. INIA Sepé shows stability of herbage accumulation and ability to adjust tiller size and population density.

## KEYWORDS

LAI, *Paspalum notatum*, Tiller density, Tiller size

## 1 | INTRODUCTION

*Paspalum notatum* (Bahiagrass) is a native type C<sub>4</sub> summer perennial grass, and one of the most frequent species in the natural fields of Uruguay noted for its high stability and the longevity of the plant population in communities (Hirata, 2004). Taking into account the potential of *P. notatum*, and to value the genetic resources of Uruguay, a national collection of germplasm was done in 2006 to characterize, value and conserve genetic resources. This process evaluated forage and seed production, adaptation to different environments, sanitary aspects, and colonizing capacity. As a result of this process, the

cultivar called INIA Sepé was selected. This grass can be classified within the functional type of resource-conserving plants (Cruz et al., 2002), according to its high tiller longevity (Giorello, 2020). For this type of forage, stable behaviours are expected in production, even when facing important changes in the environment where they develop, such as contrasting levels of nitrogenous nutrition.

Although the species is native to this region, there were very few studies that evaluated its growth, persistence, grazing management, and animal production in Uruguay and South America. At the same time, the impact of grazing on plant communities is known to determine the amount of photosynthesis that can happen as a

consequence of the removal of tissue. Modifications made in the light environment determine the patterns of competition and the morphogenic responses of each individual, and ultimately, the structure of the pasture and the growth potential (Gastal & Lemaire, 2015). In turn, forage production can be stable in a wide range of defoliation regimes (Sbrissia et al., 2018). This is possible through compensation mechanisms between the number and the size of individuals, as well as between growth, photosynthetic efficiency, and senescence (Birchman & Hodgson, 1983). Taller canopies support fewer larger tillers accumulating tissue at higher rates, while shorter canopies support a greater number of smaller tillers growing at a lower rate (Berone et al., 2007). The compensatory changes between population density and size of individuals (tillers in grasses and stems in legumes) in grazing pastures are a powerful tool to evaluate the adaptation and resistance of a plant to grazing (Hernández Garay et al., 1999; Matthew et al., 1995; Sbrissia & Da Silva, 2008; Sbrissia et al., 2001). The study of compensation relationships size  $\times$  population density of tillers has been useful to identify determining handling indicators since they are related to the limits of resistance of plants to defoliation and their productive potential (Matthew et al., 1995).

To contribute to the development of *Paspalum notatum* cv. INIA Sepé and in order to establish optimal grazing management, we considered subjecting this grass to different intensities of defoliation to evaluate its use plasticity and stability of forage production, using the size compensation mechanism by tiller population density as an evaluation tool. We hypothesized that the INIA Sepé cultivar *Paspalum notatum* is capable of maintaining dry matter production in a considerable range of defoliation intensities, and at contrasting levels of nitrogen in the plant, showing high flexibility and potential for its use as pasture as a result. Our objective is to know the plasticity limits and the pasture indicators to develop a correct grazing management.

## 2 | MATERIALS AND METHODS

The experiment was carried out from February 16, 2016, to February 28, 2019 at the Tambores Experimental Field, dependent on INIA Tacuarembó, in the Department of Tacuarembó. The coordinates of the site are 31°54'41.15"S; 56°13'39.35"W, and its altitude is 253 m.

The climate of the region of Uruguay, according to the Köppen classification, is Cfa type, with an average temperature of 18.5°C and average annual rainfall of 1294 mm. (Abal & Angelo, 2010). To maintain an adequate level of water in the soil, sprinkler irrigation was used during the entire period of the experiment, according to a water balance analysis done daily, taking into account physical data of the soil such as water retention (80 mm), infiltration speed, effective root exploration depth (40 cm) and daily climatic variables such as potential evapotranspiration and daily rainfall, as well as the applied irrigation. Climate data was collected with a Davies Pro 2 Plus weather station. (Davis., 2019) located within the experimental site 30 m away from the experiment.

The water balance analysis was done on a daily basis from Monday to Friday, and the irrigation was carried out in doses of

between 10 and 30 mm to avoid depletion below 50% of the water available in the soil and not to generate losses due to superficial runoff.

The experimental area was sown manually in December 2014, in rows 17 cm apart in soil slightly tilled with Rotovador Horticola Kubota. The soil of the experimental site is a Typical Eutric Vertisol, with a content of 39% of clay, 31% of sand, and 30% of silt. The chemical characteristics can be found in Table 1.

In November 2014 and November 2018, in order to prevent phosphorus, potassium and sulfur deficiencies, 100 units of  $P_2O_5$  as triple superphosphate, 70 units of  $K_2O$  as potassium chloride and 50 units of elemental S were applied with the commercial product Brimstone 90.

The experimental design was completely randomized with a split plot arrangement divided into two replications. In the largest plot (10 m<sup>2</sup>) a nitrogen treatment was applied as granulated urea (46-0-0) in 4 doses: 0, 100, 200 and 300 kg of nitrogen.ha<sup>-1</sup>.year<sup>-1</sup> divided into two applications, one at the beginning of the growing season during the month of October and the second at the beginning of December. On the smaller plot (2.5 m<sup>2</sup>), a cutting height treatment (at 3, 10, 17 and 24 cm) was defined and maintained by a weekly cut during the growing season using a Honda HRX 217 mower. In October 2017, each plot was raked in order to remove the dead material. For the evaluations, only the plots corresponding to the doses of 100 and 300 kg of nitrogen ha<sup>-1</sup>.year<sup>-1</sup> were used. The evaluation periods were identified as "year 1" for the date 15-March-2017, 13 months after the treatments, and "year 2" for the date 13-February-2019, 36 months after the beginning of the experiment.

### 2.1 | Forage mass, morphological and botanical composition, and nitrogenous nutrition index (NNI)

The evaluation of forage mass, botanical and morphological composition and nutritional value was carried out by extracting three samples per experimental unit, cut at ground level with electric shears powered by an internal battery and a 35  $\times$  70 cm rectangular steel frame. Once extracted, the samples were placed in plastic bags, distilled water was added with a sprayer and the bags were taken to a cold chamber until processed. This procedure was done to minimize breathing and perspiration. The material was taken to the laboratory and stored in a cold chamber for further processing. The entire sample was weighed in a fresh state. Subsamples were then separated for botanical analysis. The remaining material was weighed in a fresh state, taken to an oven to be dried for 48 h at 65°C, and weighed again. Therefore the percentage of total dry matter and the amount of forage mass were obtained by relating the percentage with the fresh weight sample. To determine the botanical and morphological composition from the samples, subsamples were selected, which were weighed in their fresh form and then manually separated into weeds, dead material, and *Paspalum notatum* (leaf, stem and inflorescence). Each fraction was sent to an oven to be dried for 72 h at 60°C and

**TABLE 1** Result of chemical analysis of the soil used in experiment I.

Depth cm	C organic %	pH (H <sub>2</sub> O)	N-NO <sub>3</sub> µg N/g	P (citric acid) µg P/g	K meq/100 g	S-SO <sub>4</sub> µg S/g	Ca meq/100 g	Mg meq/100 g	Zn mg/kg	B mg/kg
0–7.5	5.41	5.9	6.1	10.5	0.44	12.2	1.8	8.9	2.06	1.57
7.5–15	3.88	6.0	4.5	3.4	0.39	9.0	20.7	9.3	1.09	0.63

Source: Analysis Laboratory INIA La Estanzuela.

then weighed, thus obtaining the percentage of dry matter of each of the fractions and the total amount of dry matter of each fraction in the subsample, and subsequently related to the total sample. Finally, the samples were ground and then analysed with the NIR PERTEN DA 7250 equipment (Pertin, USA) in order to obtain the percentage of nitrogen in the green material (green leaves and stems), to finally calculate the Nitrogen Nutrition Index (NNI) through the calculation described in equation 1 (Lemaire et al., 2019):

Equation 1 - Calculation of the Nitrogenous Nutrition Index, according to the amount of Nitrogen (% in green tissues) and Total Green Dry Matter (ton DM.ha<sup>-1</sup>).

$$NNI = (\%N) / 3.6 \times (DM_{Green}^{-0.34}) \quad (1)$$

Where NNI is the Nitrogenous Nutrition Index, % N is the percentage of nitrogen in the green parts, and DM Green is the amount of dry matter of green material expressed in tons per hectare, and 3.6 together with  $-0.34$  are proposed coefficients for tropical species (Duru et al., 1997).

The mass of total forage and of each of the fractions expressed as kg of DM.ha<sup>-1</sup> was estimated from the ratio of the total weight and of each fraction with the area of the rectangle used to extract the samples.

## 2.2 | Population density and tiller size

To measure the population density of tillers, three counts were made per experimental unit, through observation in 20 × 70 cm frames. In each frame, the number of tillers was counted and then the number was related to the area where it was measured, obtaining the population density, which was expressed as the number of tillers.m<sup>-2</sup>.

To characterize tiller size, dry matter of green leaves, dead leaves and stem per tiller was evaluated, for which three samples of 20 tillers were collected from each experimental unit. The collection of these tillers was done with scissors at ground level. These 20 tillers were weighed fresh and the average amount of fresh matter per tiller was calculated. After weighing the material, green leaves, dead leaves and stems were separated, and each of the fresh fractions was weighed separately. They were then placed in an oven for 48 h at 65°C. The fractions of green leaves, dead leaves and stems per tiller were calculated by dividing the total dry matter obtained from each fraction by

20. The dry matter fraction per tiller was calculated by adding the total dry matter of green leaves, dead material and stems, and then dividing by 20.

## 2.3 | Leaf area index (LAI), leaf area per tiller, leaf area/volume ratio per tiller (R) and distance to the line $-3/2$

The average leaf area per tiller (cm<sup>2</sup>.tiller<sup>-1</sup>) was obtained from the separated leaves of the 3 samples of 20 tillers before being sent to the stove, which were measured with a model LI-3100 LI-COR leaf area integrator. The sum of the leaf area of each sample was divided by 20 and thus the final number of cm<sup>2</sup> of leaf.tiller<sup>-1</sup> was obtained. The leaf area index was calculated through the following equation:

Equation 2 - LAI Calculation for the value of La (Leaf area/Tiller) and TD (Tiller density) in tillers.m<sup>-2</sup>.

$$LAI = TD \times La \quad (2)$$

Where:

TD = number of tillers per m<sup>2</sup>, and La = average leaf area per tiller (m<sup>2</sup>).

The leaf area/tiller volume ratio was calculated through the following equation:

Equation 3 - Calculation of R from values of La (leaf area. tiller) raised to 3/2, and volume of tillers (m<sup>3</sup>).

$$R = (La)^{3/2} / V \quad (3)$$

Where:

La = mean leaf area per tiller (m<sup>2</sup>), and V = mean volume per tiller (m<sup>3</sup>) cleared from a constant apparent density of 950 kg.m<sup>-3</sup> (Sbrissia, 2004).

The pairs of data between mass and population density of tillers were used to generate linear regression equations on the logarithmic scale, and the distance of these points to a hypothetical line of inclination  $-3/2$  was calculated.

## 2.4 | Morphogenesis

In order to evaluate tissue flow (growth and senescence) in each treatment, 15 tillers were marked in each experimental unit, totaling

240 tillers. These were identified with white plastic rings and a plastic label was placed next to each one for identification. Evaluations were only carried out during year 2 of evaluation on the following dates: 13-February-2019; 20-February-2019; 27-February-2019 and 05-March-2019, where the leaves were numbered in each tiller and the total extended length of each leaf blade was measured from the tip to the ligule in expanded leaves and from the tip to the ligule of the last leaf expanded in the case of expanding leaves. Leaves were also classified as expanding (no visible ligule), expanded (with visible ligule or arrested growth), and defoliated (with cut parts).

Leaf expansion and senescence rates were calculated ( $\text{cm.tiller}^{-1}.\text{day}^{-1}$ ). For the purposes of converting  $\text{cm.tiller}^{-1}$  into  $\text{kg DM.ha}^{-1}$ , the tillers sampled were measured for tiller size determinations and thus generated a conversion factor ( $\text{mg.mm}^{-1}$ ) for each treatment. After obtaining the growth rate in milligrams, it was transformed into kilograms and referred to area by multiplying by the density of tillers obtained according to section Population density and tiller size.

## 2.5 | Statistical analysis

The basic assumptions of the analysis of variance, data normality and variance homogeneity were tested, assuming model additivity and error independence. When necessary, data was transformed before the analysis of variance. This was carried out with the Infostat software package (Balzarini et al., 2008). The results were analysed according to the moment of determination of the variables, in March 2017 (year 1) and in February 2019 (year 2). Height and nitrogen were considered as fixed effects and the year as a random effect. The comparison of means, when necessary, was done using the Tukey test ( $p < .05$ ). Additionally, Pearson correlations were made between the variables LAI, Distance to the line of  $-3/2$  and Growth.

In the case of the simple linear regressions carried out in the size  $\times$  population density compensation graphs of tillers, the PAST® software was used, which allows calculating the angular coefficients of the regressions by the RMA (Reduced Major Axis) method, which is recommended in situations where there could be experimental error in both axes (Matthew et al., 1995).

## 3 | RESULTS

Accumulation of forage, structural characteristics and nitrogenous nutrition index of the pasture.

Net forage accumulation varied ( $p < .05$ ) with the nitrogen dose and with the management height, with higher values being recorded with 300 compared to 100  $\text{kg N.ha}^{-1}$  ( $71.3$  and  $57.8 \pm 3.27$   $\text{kg DM.ha}^{-1}.\text{day}^{-1}$ , respectively), and less with handling height at 3 cm ( $33.0$ ,  $70.8$ ,  $75.2$  and  $79.0 \pm 4.62$  for treatments at 3, 10, 17 and 24 cm, respectively).

The forage mass varied with the nitrogen dose, with the year of evaluation, with the management height and with the interaction year of evaluation by management height ( $p < .05$ ). Higher forage mass values were recorded for the dose of 300 relative to the dose of 100  $\text{kg N.ha}^{-1}$  ( $6000$  and  $4390 \pm 295$   $\text{kg DM.ha}^{-1}$ , respectively). In general, higher forage mass values were recorded with higher management heights, and in year 2 the values for the 10 and 24 cm treatments were lower than in year 1 (Table 2).

The percentage of *Paspalum* leaves varied ( $p < .05$ ) with the year of evaluation and with the nitrogen doses, being higher in year 2 compared to year 1 ( $50.0$  and  $27.5 \pm 1.10\%$ , respectively). On the other hand, a higher value was recorded with the 100 nitrogen dose compared to the 300  $\text{kg N.ha}^{-1}$  dose ( $40.5$  and  $37.0 \pm 1.10\%$ , respectively).

The percentage of *Paspalum* stems varied ( $p < 0.05$ ) with the handling height, the evaluation year, with the nitrogen doses and with the interaction year of evaluation by handling height, with a higher value being recorded with the 300 nitrogen dose compared to the 100  $\text{kg N.ha}^{-1}$  ( $27.1$  and  $17.9 \pm 1.52\%$ , respectively). In year 1, there was no difference between handling heights, but in year 2 the 3 cm treatment presented a lower percentage of stems than the 10, 17 and 24 cm treatments (Table 3).

The percentage of inflorescences during the experiment was extremely low, not exceeding (except for one occasion) 1% of the total forage mass. Similarly, the percentage of weeds in the forage mass was also reduced (less than 5%), and varied with the year of evaluation, that was higher in year 2 compared to year 1 ( $4.0$  and  $0.7 \pm 0.81\%$ , respectively).

The percentage of dead material varied ( $p < .05$ ) with the handling height, evaluation year and with the interaction handling height by evaluation year. In year 1, the percentage of dead material increased with higher handling heights, but during year 2 the highest value was recorded for the 3 cm treatment relative to the others (Table 4). In general, the percentage values of dead material were lower in year 2 compared to year 1.

The Leaf Area Index (LAI) varied ( $p < .05$ ) with the nitrogen dose, handling height and with the interaction year of evaluation by handling height. Higher values were recorded for the 300 dose compared to 100  $\text{kg N.ha}^{-1}$  ( $2.61$  and  $2.19 \pm 0.098$ , respectively). In general, higher values were recorded with higher handling heights, and there was a difference in LAI for the same handling height only for the 10 cm treatment, with a higher value recorded in year 1 compared to year 2 (Table 5).

The Nitrogenous Nutrition Index varied ( $p < 0.05$ ) with the handling height, nitrogen dose and year of evaluation. There was a difference between the 3 and 24 cm treatments, with intermediate (and similar) values for the 10 and 17 cm treatments ( $0.67$ ,  $0.76$ ,  $0.81$  and  $0.90 \pm 0.033$  for treatments at 3, 10, 17 and 24 cm, respectively). In relation to the nitrogen doses, higher values were recorded for the 300 dose compared to 100  $\text{kg N.ha}^{-1}$  ( $0.90$  and  $0.70 \pm 0.033$ , respectively). In relation to the year of evaluation, higher values were recorded during year 2 relative to year 1 ( $0.91$  and  $0.66 \pm 0.032$ , respectively).

**TABLE 2** *Paspalum notatum* INIA Sepé forage mass maintained at contrasting handling heights between March 2017 and February 2019.

	Height (cm)									
Year	3 (kg DM.ha <sup>-1</sup> )		10 (kg DM.ha <sup>-1</sup> )		17(kg DM.ha <sup>-1</sup> )		24(kg DM.ha <sup>-1</sup> )		Mean	SE
1	1500	Ac	5460	Ab	6620	Ab	10,150	Aa	5930 A	485
2	1320	Ac	4120	Bb	5690	Aab	6650	Ba	4450 B	522
Median	1410	c	4790	b	6150	b	8400	a		
S.E.	136		295		456		839			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

**TABLE 3** Percentage of stems in the forage mass of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights between March 2017 and February 2019.

	Height (cm)									
Year	3 (%)		10 (%)		17 (%)		24 (%)		Median	SE
1	20.0	Aa	14.3	Ba	19.3	Ba	19.5	Ba	18.0 B	2.68
2	7.3	Ab	30.8	Aa	33.3	Aa	35.3	Aa	26.6 A	3.35
Median	13.6	b	22.5	a	26.3	a	27.4	a		
SE	3.84		2.41		2.22		3.37			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

**TABLE 4** Percentage of dead material in the forage mass of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights between March 2017 and February 2019.

Year	Height (cm)								Median	SE
	3 (%)		10(%)		17 (%)		24 (%)			
1	46.0	Ab	55.3	Aab	56.5	Aa	48.8	Aab	51.6 A	2.27
2	38.0	Aa	15.5	Bb	13.3	Bb	8.5	Bb	18.8 B	2.75
Median	42.0	a	35.4	ab	34.9	Ab	28.6	ab		
SE	4.11		1.36		2.27		1.26			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

**TABLE 5** Leaf Area Index (LAI) of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights between March 2017 and February 2019.

Year	Height (cm)								Median	SE
	3	10	17	24						
1	1.60	Ab	2.90	Aa	2.45	Aab	3.23	Aa	2.54 A	0.195
2	0.88	Ac	1.80	Bb	2.80	Aa	3.53	Aa	2.25 A	0.185
Median	1.24	c	2.35	b	2.63	b	3.38	a		
SE	0.172		0.177		0.236		0.168			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

### 3.1 | Population density and individual characteristics of tillers

The population density of tillers varied ( $p < 0.05$ ) in the year of evaluation, the handling height and the interaction dose of nitrogen by handling height and by year of evaluation. In general, higher densities were recorded for the lower handling heights, except in year 2 for the dose of 100 kg N.ha<sup>-1</sup>, when there was no difference between handling heights (Table 6).

The size of tillers varied ( $p < 0.05$ ) with the nitrogen dose, evaluation year, handling height, evaluation year by nitrogen dose interaction, and evaluation year by handling height interaction. The tillers were higher for the treatment 300 kg N.ha<sup>-1</sup> in year 2 of evaluation (Table 7). In general, higher values were recorded with higher handling heights, and the difference in mass for the same height was recorded only for the 24 cm treatment, which presented a higher value in year 2 compared to year 1 (Table 8).

**TABLE 6** Population density of tillers of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights and fertilized with two doses of nitrogen between March 2017 and February 2019.

Nitrogen (kg.Ha <sup>-1</sup> )	Height (cm)								Median	SE
	3 (tillers.m <sup>-2</sup> )		10 (tillers.m <sup>-2</sup> )		17 (tillers.m <sup>-2</sup> )		24 (tillers.m <sup>-2</sup> )			
Year 1										
100	2848	Aa	1694	Aab	1181	Ab	946	Ab	1668 A	225
300	2350	Aa	1939	Aa	1357	Ab	1327	Ab	1743 A	78
Median	2599	a	1817	b	1269	c	1137	c		
SE	269		133		97					
Year 2										
100	1635	Aa	1123	Aa	1032	Aa	848	Aa	1159 A	149
300	2011	Aa	1095	Ab	1071	Ab	899	Ab	1269 A	72
Median	1823	a	1097	b	1064	b	873	b		
SE	220		54		52		26			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

Year	Nitrogen (kg.Ha <sup>-1</sup> )				Median (g. tiller <sup>-1</sup> )	SE
	100 (g. tiller <sup>-1</sup> )	300 (g. tiller <sup>-1</sup> )				
1	0.25	Aa	0.26	Ba	0.26 B	0.016
2	0.27	Ab	0.36	Aa	0.31 A	0.016
Median	0.26	b	0.31	a		
SE	0.021		0.008			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

Year	Height (cm)				Median	SE
	3 (g. tiller <sup>-1</sup> )	10 (g. tiller <sup>-1</sup> )	17 (g. tiller <sup>-1</sup> )	24 (g. tiller <sup>-1</sup> )		
1	0.08	Ac	0.21	Ab	0.33	Aa
2	0.06	Ac	0.26	Ab	0.36	Ab
Median	0.07	d	0.24	c	0.34	b
SE	0.013		0.015		0.032	0.033

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

Year	Height (cm)				Median	SE
	3 (cm <sup>2</sup> )	10 (cm <sup>2</sup> )	17 (cm <sup>2</sup> )	24 (cm <sup>2</sup> )		
1	5.9	Ac	15.9	Ab	18.9	Ab
2	4.4	Bd	16.4	Ac	26.3	Ab
Median	5.3	d	16.3	c	22.6	b
SE	0.25		0.75		2.02	1.15

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.

Leaf area per tiller varied ( $p < 0.05$ ) with the year of evaluation, the handling height and the interaction year of evaluation by handling height. In general, higher values were recorded for higher handling

heights, but there was a difference between years for the same handling height for treatments at 3 and 24 cm, with lower and higher values in year 2 for treatments at 3 and 24 cm, respectively (Table 9).

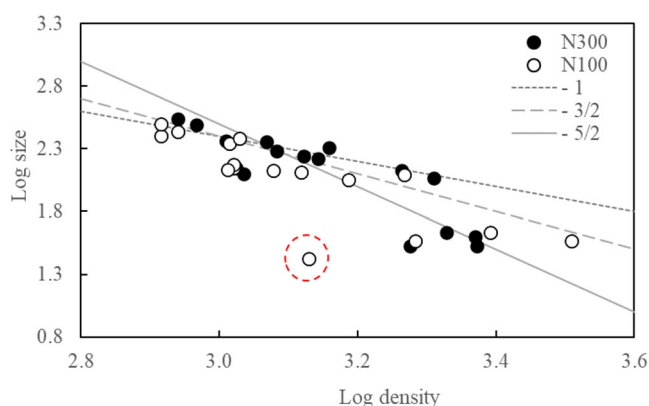
**TABLE 7** Size of tillers of *Paspalum notatum* INIA Sepé fertilized with two doses of nitrogen between March 2017 and February 2019.**TABLE 8** Size of tillers of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights between March 2017 and February 2019.**TABLE 9** Leaf area per tiller of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights in March 2017 and February 2019.



**TABLE 10** Leaf area:Volume of tillers ratio of *Paspalum notatum* INIA Sepé maintained at contrasting handling heights between March 2017 and February 2019.

Year	Height (cm)								Median	SE
	3	10	17	24	24	24	24	24		
1	18.5	Ac	28.2	Aab	24.0	Bbc	35.0	Aa	26.4 B	1.85
2	14.7	Ac	25.0	Ab	35.7	Aa	42.7	Aa	29.6 A	1.92
Median	16.6	c	26.6	b	29.9	b	38.9	a		
SE	2.72		1.19		1.21		1.98			

Note: Means with a common uppercase letter in the column or lowercase in the row are not significantly different ( $p > .05$ ). SE indicates the Standard Error of the variable.



**FIGURE 1** Regression between size of tillers logarithm and population density of tillers logarithm of tillers of *Paspalum notatum* cv. INIA Sepé.

**TABLE 11** Regression parameters between Size Logarithm and Population Density Logarithm of tillers, Ca, Cr and predicted slope of the line according to nitrogen dose and evaluation year.

Parameters	Regression	Ca	Cr	Predicted slope
100 kg N.ha <sup>-1</sup> - Year 1				
Intercept	6.97 (0.62)			
Slope	-1.54 (0.2)	-0.85	0.7	-1.65
R <sup>2</sup>	0.90			
p	0.00022			
300 kg N.ha <sup>-1</sup> - Year 1				
Intercept	10.68 (1.64)			
Slope	-2.68 (0.51)	-2.33	1.43	-2.40
R <sup>2</sup>	0.78			
p	0.002			
100 kg N.ha <sup>-1</sup> - Year 2				
Intercept	10.14 (0.85)			
Slope	-2.62 (0.28)	-2.77	1.74	-2.53
R <sup>2</sup>	0.94			
p	0.0002			
300 kg N.ha <sup>-1</sup> - Year 2				
Intercept	10.32 (1.13)			
Slope	-2.65 (0.37)	-2.59	1.48	-2.61
R <sup>2</sup>	0.89			
p	0.0003			

**TABLE 12** Pearson correlation coefficient values between LAI and Distance to the line of  $-3/2$  in year 1, and between LAI, Distance and Net forage accumulation for year 2.

Variable (1)	Variable (2)	Pearson	p-value
Year 1			
LAI	Distance	0.83	0.0001
Year 2			
LAI	Distance	0.90	0.0001
LAI	Net forage accumulation	0.93	0.0001
Distance	Net forage accumulation	0.88	0.0001

Leaf area: Volume of tillers ratio (R) varied ( $p < 0.05$ ) with the handling height, year of evaluation and the interaction handling height by year of evaluation. In general, higher values were recorded with higher handling heights, and in year 2 the value recorded for the treatment at 17 cm was higher compared to year 1 (Table 10).

Relations between size and population density of tillers and forage net accumulation.

The regressions between size logarithm and population density logarithm of tillers generated slopes close to  $-5/2$ , except for year 1 with the 100 kg N.ha dose<sup>-1</sup>, which was close to  $-3/2$ . In all cases, close values were found between the regression calculated between logarithms and the one predicted using the values of the corrections by LAI and R (Ca and Cr) proposed by Matthew et al. (1995).

The distance of the data pairs (size log  $\times$  density log) to a hypothetical line with slope  $-3/2$  varied ( $p < 0.05$ ) with the nitrogen dose, evaluation year and handling height. Higher values were recorded for a 300 dose compared to 100 kg N.ha<sup>-1</sup> (0.47 and 0.42  $\pm$  0.01, respectively), in year 1 compared to year 2 (0.51 and 0.39  $\pm$  0.01, respectively), and less in the treatment at 3 cm (0.36, 0.46, 0.47 and 0.50  $\pm$  0.02 for treatments at 3, 10, 17 and 24 cm, respectively).

A point was identified where the compensation between size and population density of tillers did not have an effect, evidenced by the distance from the regression lines, a situation recorded in an experimental unit managed with doses of 100 kg N.ha<sup>-1</sup> and 3 cm of cutting height in year 2 of evaluation (Figure 1).

There was a positive correlation between LAI and Distance to the line of  $-3/2$  in the 2 years of evaluation, as well as between Distance to the line of  $-3/2$  and net accumulation of forage in year 2 (Table 12).

## 4 | DISCUSSION

The net accumulation of forage did not vary in a wide range of defoliation intensities, being lower with a handling height of 3 cm, regardless of the dose of nitrogen fertilization. This forage, from a group of resource-conserving plants, does not maximize capture mechanisms and therefore shows great stability in terms of its productive behaviour. Although there were differences in tiller populations, these being smaller as management heights increased, clearly, the net accumulation per tiller must have increased at similar rates, so that the net accumulation of forage did not vary between 10 and 24 cms. In the case of the 3 cm handling height, the reduction in leaf area probably did not allow the capture of enough energy to maintain high growth rates.

In turn, higher handling heights generated greater LAI and greater forage mass (Table 2), as a result of the greater opportunity for tissue accumulation caused by less severity in the cuts made at higher heights. A greater LAI and greater forage mass at higher altitudes and a stable net accumulation of forage should be caused by a lower photosynthetic efficiency of the canopies managed at higher altitudes. In turn, the morphological composition was relatively stable, characterized by a small variation in the percentage of leaves, stems, and dead material, except for the handling height at 3 cm in year 2 (Tables 2, 3, and 4), which resulted in a lower percentage of stems compared to the remaining handling heights. This fact is consistent with the condition of a *steady state* generated by experiments of this nature in which plants are subjected to defoliation regimes with strict control of handling height (Sbrissia et al., 2010; Sbrissia et al., 2020). This characteristic is a positive aspect given the possible flexibility in grazing management that it allows, since the greater presence of stems is associated with a lower destination of carbohydrates to form leaves, which in turn determines a lower photosynthetic efficiency. Productive stability is manifested not only in the value of net accumulation but also in the stability of the components of the accumulated forage. It is important to mention that in this experiment, weekly machine cuts to maintain handling heights could have minimized the differences in the percentage of stems between handling heights, as well as reduced the presence of inflorescences, a percentage which was in general very low and only different in height at 3 cm in evaluation year 1. In the remaining situations, the percentage of inflorescences did not exceed 1% of the total forage mass. By maintaining a very high frequency of defoliation, there was a very important decapitation of reproductive tillers before the visualization of inflorescences, which resulted in a low percentage in the forage mass. Although the percentage of weeds was different between years of evaluation, it remained at very low levels. This reduced presence of weeds is associated with the dense cover of *Paspalum notatum* and its great competitive ability. In evaluation year 1, the percentage of dead material increased with higher handling heights, different from evaluation year 2, where the accumulated dead material was removed. The removal of dead material may have caused differences in nitrogen dynamics, with differences in NNI values being observed, which were higher in evaluation year 2 and higher at higher handling heights.

Regarding nitrogen doses, the dose of 300 kg N.ha<sup>-1</sup> generated a higher LAI and greater forage mass, determined by a larger tiller size, despite not finding significant differences in tiller population density and individual leaf area compared to the dose of 100 kg N.ha<sup>-1</sup>.

Higher handling heights resulted in pastures with a lower population density of large tillers, with greater individual leaf area relative to lower handling heights (Tables 6 and 8). The reduction in population density was less than the increase in leaf area per tiller, which resulted in the highest values of forage mass and LAI in the pastures managed with the highest cutting heights. These results clearly indicate an inverse relationship between number and size of tillers, a pattern described for a series of other forage species of temperate (Hernández Garay et al., 1997; Hernández Garay et al., 1999) and tropical (Sbrissia et al., 2001, 2003, 2018) climate, and that characterizes the mechanism known as tiller population size by density compensation. This mechanism, in certain situations, allows the LAI of the pasture to be kept relatively stable within a range of handling heights through variation in size and number of tillers (Matthew et al., 1995), and can characterize the amplitude of management flexibility of a given species. Within the optimal handling scope, reductions in tiller size (and leaf area) are offset by increases in population density. From the moment where reductions in tiller size (shorter handling height) are no longer compensated by the proportional increase of population density, the LAI is reduced, foliage accumulation is lower and an emergence of invasive plants can be observed. This compensation relationship is characterized by a linear relationship between the logarithm of the mass per tiller and the logarithm of the population density of tillers with an angular coefficient of  $-3/2$  (integral compensation) or  $-5/2$  (partial compensation and LAI increasing with handling height) (Sackville Hamilton et al., 1995). In this study, an angular coefficient of the linear relationship close to  $-2.5$  was observed, a condition corresponding to the segment of the multiphase diagram Sackville Hamilton et al. (1995), which corresponds to self-compensation at a rate of  $-5/2$  with LAI and variable R (Tables 5 and 10). This situation was different only in evaluation year 1 with the 100 kg N.ha dose<sup>-1</sup>, where the recorded slope was close to  $-3/2$ . This difference may be due to the limitation in the production potential imposed by the low dose of nitrogen, which did not allow the production of a greater amount of tissues, mainly leaves, given the restriction generated by the competition with the decomposition process of the dead material present, and the low dose provided in the treatment. This circumstance possibly determined a low production potential, because, due to a nutritional deficiency, it was not able to continue increasing its LAI in order to be able to capture a greater amount of light, stabilizing the leaf area index and compensating the size and density of the tillers at a rate of  $-1.5$ . In evaluation year 2 (third of the experiment) a plot was unable to compensate for the reduction in size of its tillers with the increase in population density of the same events, evidenced in Figure 1, where the distant location of the different straight lines of compensation is clearly observed (treatment 3 cm with 100 kg N.ha<sup>-1</sup>). On the other hand, the model proposed by Sackville-Hamilton et al. (1995), that foresees a straight line with slope  $-3/2$  when the difference in LAI



and R is zero with the variation in pasture height, also considers the possible effect of apparent density, although given the difficulty of estimating it and the low variability in the characteristic, it is considered constant over a wide range of handling heights. The adjustments made considering LAI and R, called Ca and Cr, respectively (Matthew et al., 1995), foresee corrections in the cases where LAI is greater due to managing the pastures at a greater height, and therefore the line should be dislocated to a position with larger intercepts. For this to happen there should be a theoretical increase in tiller density, and this is what the correction shows. As per Cr, it would be the case of variations in the shape of the tillers, for which greater variation in the shape that was not coincident with the variation in LAI would generate predictions greater than  $-3/2$ . In the four cases analysed corresponding to the two doses of nitrogen in each year of evaluation, the lines predicted by calculating Ca and Cr (Table 11) coincide with the values observed by regressing the tiller size logarithm and the population density logarithm. When the values of Ca and Cr were compared, a higher value was observed in the case of Ca, indicating a greater effect of variation caused by the LAI compared to the effect caused by variations in the shape of the tillers, and therefore recorded in variations in R. In turn, the values of R found were higher than those found for *Cynodon* by Sbrissia et al. (2001), and lower than those found in *Brachiaria* (Sbrissia & Da Silva, 2008) and Ryegrass (Hernández Garay et al., 1999), although similar to this last species in that the value increased with higher handling heights, which indicates that the plant grows proportionally producing a greater number of leaves compared to the number of stems, a fact that differentiates this species from the majority of tropical species, allowing for greater flexibility in grazing management. In turn, lower values of R could be associated with plants with resistance to different stresses (Matthew et al., 2000), given the lower leaf area: volume ratio, which could be a way to avoid loss of water.

On the other hand, Matthew et al. (1995) propose that the distance to the hypothetical line of  $-3/2$  of any point in the regression log size and log density of tillers would be an indicator of pasture productivity, with a high and positive relationship between distance to the line and LAI. This was confirmed in the present work (Table 12), as well as in previous works on temperate species by Hernandez Garay et al. (1997, 1999), and in tropical species by Sbrissia et al. (2001, 2003). Unlike previous works, net forage accumulation was correlated with LAI and distance, also resulting in high and positive correlations ( $p < .0001$ ), reaffirming the value of the distance to the line as a predictive value both of LAI and mainly of pasture forage production.

## 5 | CONCLUSIONS

Similarly, to other grasses of temperate and tropical climate, *Paspalum notatum* cv INIA Sepé presents great plasticity and management flexibility characterized by the stability of forage production and its ability to make adjustments in the size and population density of tillers from 10 to 24 cm of handling height.

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## CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest.

## DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on request from the corresponding author. The data are not publicly available due to privacy or ethical restrictions.

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