

Production and morphogenetic traits of *Megathyrus maximus* "Aruana" with nitrogen fertilization in silvopastoral and full sun systems

Características produtivas e morfofisiológicas de *Megathyrus maximus* "Aruana" com adubação nitrogenada em sistema silvipastoril e pleno sol

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Highlights

Nitrogen fertilization increased herbage quality by increasing crude protein content.
The presence of light influenced the morphological development of the forage.
The higher leaf area index influenced dry matter production and herbage accumulation.

Abstract

The objective of this study was to assess the impact of nitrogen fertilization on *M. maximus* cv. Aruana cultivated in silvopastoral and full-sun systems, focusing on morphogenetic, structural, and productive traits, as well as chemical composition. Silvopastoral systems promote productive efficiency and sustainability in animal husbandry, leading to improved herbage quality and enhanced thermal comfort for the animals. Nitrogen fertilization affects the growth of tropical forages. The study evaluated the

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system used (silvopastoral or full sun), the presence of fertilization (with or without), and distinct intervals (21, 42, 63, or 84 days) on productive and morphogenetic traits of the forage. The silvopastoral system exhibited increased canopy height, while the full-sun condition favored dry matter production and animal weight gain. Fertilization benefited basal tiller density and morphogenesis. The crude protein content was higher in full sun. Full sun displayed benefits in terms of both herbage production and composition, whereas the silvopastoral system excelled particularly in terms of canopy height. Fertilization improved several traits, fostering tissue turnover. The study highlights the importance of proper management in silvopastoral systems to optimize herbage production and quality.

Key words: Morphogenesis. Mineral fertilization. Shading. LAI.

Resumo

O objetivo do presente trabalho foi avaliar o efeito da adubação nitrogenada em *M. maximus* cv. Aruana cultivado em sistemas silvipastoril e pleno sol sobre as características morfológicas, estruturais, produtivas e composição química. Sistemas silvipastoris promovem eficiência produtiva e sustentabilidade na produção animal, melhorando a forragem e o conforto térmico. A adubação nitrogenada afeta o crescimento de forrageiras tropicais. O estudo avaliou o sistema utilizado (silvipastoril e pleno sol), adubação (com e sem) e períodos (21, 42, 63 e 84 dias) em características produtivas e morfológicas da forrageira. O sistema silvipastoril elevou a altura do dossel, enquanto o pleno sol favoreceu a matéria seca e ganho de peso animal. A adubação beneficiou a densidade de perfilhos basais e morfogênese. O teor de proteína bruta foi maior no pleno sol. Pleno sol teve vantagens na forragem e composição, enquanto o silvipastoril destacou-se na altura do dossel. A adubação melhorou várias características, incentivando renovação tecidual. O estudo destaca a importância do manejo adequado em sistemas silvipastoris para otimizar produção e qualidade forrageira.

Palavras-chave: Morfogênese. Adubação mineral. Sombreamento. IAF.

Introduction

Silvopastoral systems have demonstrated a remarkable capacity to enhance the productive efficiency of their components, significantly contributing to the sustainability of animal production (Paciullo et al., 2017; Anjos & Chaves, 2021). Moreover, these systems increase soil fertility, improve herbage quality and production, and enhance the thermal comfort of animals.

They also serve to rehabilitate degraded areas, sequester carbon, and bolster biodiversity, generating diversified income streams for producers (Abraham et al., 2014; Paciullo et al., 2014; López-Carrasco et al., 2015). However, the management of light competition within silvopastoral systems assumes great importance to avert potential limitations on forage potential due to shading caused by trees (Neel et al., 2016; Santiago-Hernández et al., 2016).

Irrespective of the chosen system, nitrogen (N) deficiency can severely impact the growth of tropical forages (Silveira et al., 2015). The application of N during the vegetative phase of plants leads to heightened shoot growth, improved light uptake, augmented photosynthetic activity, and increased nutrient uptake through roots (Fagan et al., 2016). In shaded environments, the availability of soil N tends to be more favorable due to a gradual reduction in soil moisture, fostering microbial activity and mineralization processes (Araújo et al., 2017).

To harness the benefits of both shading and fertilization, the selection of a suitably adaptable and widely distributed forage species is imperative. One such example are *Megathyrsus maximus* cultivars. The attributes of moderate height and reduced stem length render *M. maximus* cv. Aruana a highly promising option for sheep grazing (Sacramento et al., 2019). With its high potential for generating dry matter per unit area, adaptability, nutritional excellence, and ease of establishment, *M. maximus* stands out as one of the best tropical forage choices (Lima Veras et al., 2020).

The experiment aimed to validate the hypothesis that the combination of silvopastoral practices with N fertilization exerts a positive influence on forage plants, thereby fostering their growth,

development, and chemical composition. The objective encompassed the evaluation of morphogenetic, structural, and productive traits as well as the chemical composition of *M. maximus* cultivated in a silvopastoral system or in full-sun conditions, with and without nitrogen fertilization, and grazed by sheep.

Material and Methods

Location

The study was conducted in a field setting at the Sheep and Goat Farming Teaching and Research Unit of the Federal Technological University of Paraná, Dois Vizinhos campus, from December 2019 to March 2020. Situated in the southwest region of Paraná, the institution occupies a latitude of 25° 42' S and longitude of 53° 03' W, with an approximate elevation of 520 m above sea level. The soil type is categorized as an Oxisol with a clayey texture (Empresa Brasileira de Pesquisa Agropecuária [EMBRAPA], 1999). The region is characterized by a mesothermal humid subtropical climate (Cfa), characterized by an average annual precipitation of 1,953 mm and mean annual temperatures of 25.2 °C (maximum) and 14.7 °C (minimum) (Alvares et al., 2013) (Figure 1).

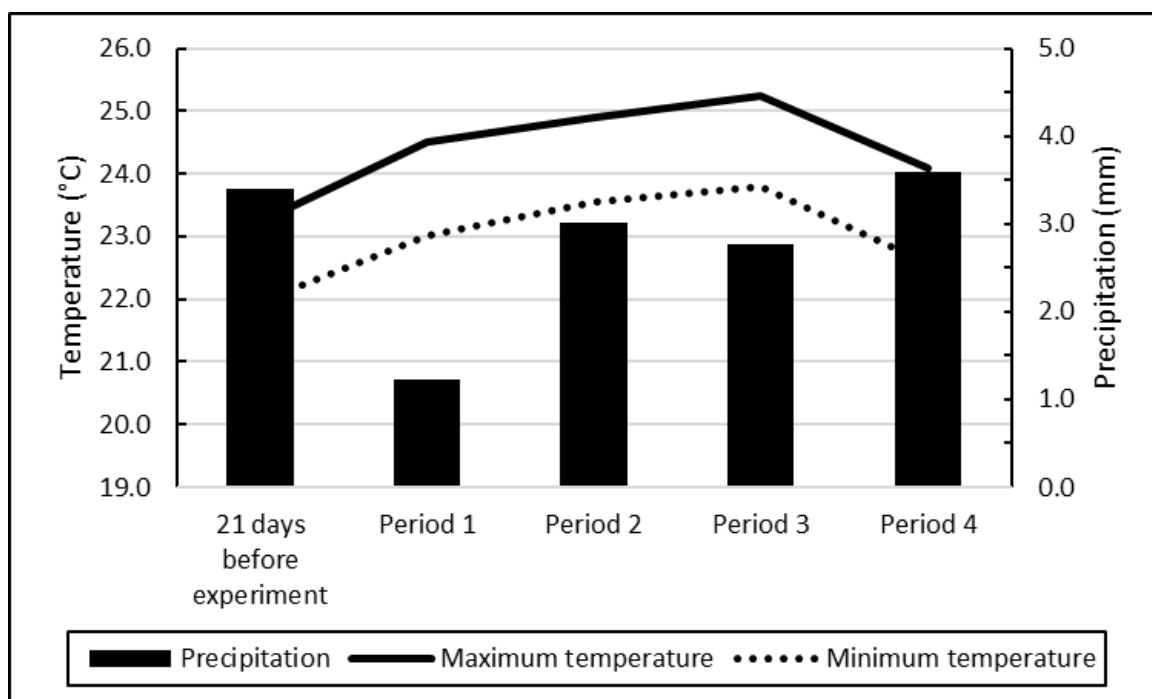


Figure 1. Meteorological data [minimum and maximum temperatures (°C) and precipitation (mm)] observed during field evaluations (November 2019 to March 2020). Source: GPCS (2020).

Experimental procedures

This study received approval from the Committee on Ethics in the Use of Animals (CEUA - Universidade Tecnológica Federal do Paraná - approval no. 2019-34), in compliance with the guidelines established by the National Council for the Control of Animal Experimentation (CONCEA).

The experimental treatments encompassed the following scenarios: Aruana pasture under full sun with nitrogen fertilization; Aruana pasture under full sun without nitrogen fertilization; Aruana pasture in a silvopastoral system with nitrogen fertilization; and Aruana pasture in a silvopastoral system without nitrogen fertilization. Nitrogen fertilization was applied

in the treatment paddocks with agricultural urea (46% total N) as a single application, dosed at 200 kg N ha⁻¹. This application occurred after the leveling cut of the plots to optimize the efficiency of fertilization, approximately 15 days before the initiation of the experimental assessments.

The study area spanned a total of 0.48 ha, cultivated with *M. maximus* cv. Aruana. This space was divided into two distinct systems: six paddocks within the silvopastoral system (each with an area of 400 m²) comprising *Cordia trichotoma* and *Peltophorum dubium* trees, and six paddocks exposed to full sun (each with an area of 400 m²). The pasture was established in 2010, while the tree components were introduced in the years 2013/2014 within the

silvopastoral system. The tree arrangement extended in an East-West direction, with trees placed in four double rows at the ends of each paddock. The spacing between trees was set at 10 m, with a planting distance of 2.00 m between individual plants and 1.50 m between rows. According to a forest survey conducted in 2019, the average crown diameter of *C. trichotoma* specimens was 3.05 m, with a crown height of 3.10 m and an average height of 7.90 m. For *P. dubium*, data collected in 2018 indicated an average crown diameter of 2.20 m, crown height of 2.00 m, and an overall height of 3.90 m. On average, each tree provided a shade area spanning 36 m² (Cipriani et al., 2016). Both systems were equipped with drinkers and troughs to facilitate mineral supplementation for the animals.

Before initiating the experiment, the overall soil pH of the designated area underwent correction using dolomitic limestone (RNV 95.2%) with the intention of raising the base saturation to 70% (BS: 55.9%).

Each experimental phase spanned 21 days, and the entire experiment comprised four of these phases, totaling an evaluation period of 84 days. Twenty-four male lambs (Dorper × Santa Inês), with an average age of 60±15 days and an average weight of 23±3.5 kg, were employed. Continuous grazing with a variable stocking rate was employed using the “put and take” technique (Mott & Lucas, 1952). The adaptation period was accounted for within the experimental phases since the animals used for the evaluations had prior exposure to a semi-intensive rearing system, implying that grazing was already a familiar activity.

Body weight gain per hectare (BWG ha⁻¹) was calculated utilizing the average daily gain (ADG) determined for each assessment period (21 days). This was computed using the equation:

$$\text{BWG} = (\text{ADG} \times \text{Number of days in the paddock}) / \text{Paddock area.}$$

Pasture evaluations were carried out at 21-day intervals, maintaining a consistent supply of 10% herbage mass (HM) (10 kg DM 100 kg BW⁻¹ animal⁻¹ day⁻¹). The available HM was determined by employing the double-sampling method (Wilm et al., 1994).

Morphogenesis

For the determination of morphogenetic and structural characteristics, three transects, each featuring five tillers identified by colored wires, were utilized within each of the experimental paddocks, as by Carrère et al. (1997). The transects were distributed within the paddocks to ensure representative sampling. Evaluations occurred twice weekly, with tillers being replaced at the start of a new period to maintain representativeness.

Measurements were conducted using a millimeter-graded ruler, capturing the following variables: leaf and stem lengths, the number of mature leaves along with their dimensions (in centimeters), classification based on leaf status (senescent or not) and whether they were intact or defoliated, following Lemaire and Chapman (1996). From this data, the following parameters were calculated: Leaf Appearance Rate: ratio of fully expanded leaves to the number of days

in the experimental period; Leaf Elongation Rate: difference between final and initial leaf blade lengths divided by the number of days in the period; Stem Elongation Rate: difference between final and initial stem lengths divided by the number of days in the period; Leaf Senescence Rate: sum of senescent leaf blade lengths divided by the number of days in the period; Number of Live Leaves (NLL): average count of fully expanded leaves without signs of senescence; Final Leaf Length; and Leaf Lifespan (LLS), estimated using the equation $LLS = NLL \times \text{Phyllochron}$.

Herbage mass and tiller density

To ascertain the HM (kg ha^{-1}), plant material was collected from each of the experimental paddocks utilizing the double-sampling technique (Wilm et al., 1994). This involved visually estimating 12 points and performing three real estimates for each experimental paddock. The herbage was cut near the ground using a square with an area of 0.25 m^2 at the real points. The structural composition of the total collected material was determined. Basal and aerial tiller density were gauged using a 0.0625-m^2 square at the beginning of each experimental period. Tiller counts within this area were conducted, with three assessments performed per paddock to ensure the representation of each area.

Daily accumulation rate and leaf area index

To establish the daily herbage accumulation rate (DAR), grazing exclusion cages measuring 0.25 m^2 were installed in each of the experimental paddocks. One

cage was positioned in a representative area, while a similar point was selected as a comparison. Herbage available within both cages was cut using a 0.25-m^2 square. The collected samples were dried in an oven at $55 \text{ }^\circ\text{C}$ for 72 h, weighed, and DAR was derived from the weight discrepancy between the two samples, divided by the number of days in the evaluation period, following Klingman et al. (1943).

For the determination of the leaf area index (LAI), plant material was collected from each experimental paddock within a 0.25-m^2 area, chosen for its representativeness. The collected material was sorted into leaves, stems, and dead material. Within each plot, leaf blades from approximately 10 tillers were used to compute LAI, based on methodologies outlined by Zanchi et al. (2009) and Sousa et al. (2015). The remaining material was weighed and dried in a forced-air oven ($55 \text{ }^\circ\text{C}$) to determine the DM content.

Leaf area (LA) was computed using the AFUFT method (Sousa et al., 2015) with a millimeter-graduated ruler, employing measurements of each sampled leaf (total length: measurement A; width of the blade base: measurement B; width of the middle of the blade: measurement C, all in cm). The calculations included:

$$\text{LA} = \text{Triangle area } [((\text{Measurement C}) \times (\text{Measurement A}/2))/2] + \text{Trapezium area } [((\text{Measurement C} + \text{Measurement B})/2) \times (\text{Measurement A}/2)]; \text{ and}$$

$$\text{LAI} = (\text{Leaf weight})/(\text{Leaf weight} + \text{Stem weight}) \times \text{Dead material weight} \times \text{LA}/10000.$$

Chemical analysis

To determine the chemical composition, samples of leaves and stems were collected, dried in a forced-air oven (55 °C) for 72 h, and then ground using a Wiley mill with a 2-mm sieve. Dry matter (method 934.01) and crude protein (method 981.10) were determined following Association of Official Analytical Chemists [AOAC] (2016) protocols. Neutral detergent fiber and acid detergent fiber were gauged in accordance with Van Soest et al. (1991), using filter bags (Komareck, 1993), within an autoclave at 110 °C for 40 min (Senger et al., 2008). The lignin content was determined by treating the samples with 72% sulfuric acid (AOAC, 2016).

Experimental design and statistical analysis

The experimental design employed was completely randomized, utilizing a double factorial arrangement (presence or absence of nitrogen fertilization and two systems - full sun and silvopastoral) with repeated measures over time. The collected data underwent a normality test (Shapiro-Wilk) via the UNIVARIATE procedure. Subsequently, a two-way analysis of variance (ANOVA) with repeated measurements in time (systems, fertilization, and four collection periods: 21, 42, 63, and 84) was conducted using the PROC GLIMMIX command within the statistical software SAS® University Edition (2017). Among the considered error structures, the Components of Variation error (VC) demonstrated the best fit according to the Akaike Information Criterion (AIC) and the Bayesian Information System (BIC).

Significant results for isolated effects were subjected to mean comparison between production systems and fertilization options using the F-test. Regression analysis was also applied to assess the effect of periods, using the PROC GLIMMIX and PROC REG commands. A significance level of $\alpha = 0.05$ was adopted for all hypothesis tests.

The mathematical model below was applied to analyze the variables:

$$Y_{ijk} = \mu + T_i + R_k + P_j + TP_{ij} + e_{ijk}$$

where Y_{ijk} : observation related to production system i , fertilization option k , and period j ; μ : overall mean; T_i : effect corresponding to production system i (silvopastoral or full sun); R_k (T_i): effect corresponding to fertilization option k (with or without); P_j : effect corresponding to period j (21, 42, 63, or 84 days); TP_{ijk} : interaction effect between production system i , fertilization option k , and period j ; e_{ijk} : random error associated with production system i , fertilization option k , and period j .

For the basal tiller density variable, the PROC GLM command was employed, fitting the data with the Gamma distribution and Log link function. For the leaf senescence rate variable, the GLM was adjusted utilizing the Exponential Distribution and Logarithmic Link function. Additionally, the GLM was applied to the basal tiller density variable, using negative binomial distribution and the Log link function. The determination of model fit was based on the AIC and graphical analysis of residual adherence.

Results and Discussion

Morphogenetic and structural traits

Leaf senescence rate was not influenced ($P > 0.05$) by the evaluated treatments. Canopy height exhibited a higher value ($P = 0.0002$) in the silvopastoral system (27.52 cm) compared to full sun conditions (21.97 cm). The rates of leaf appearance, as well as leaf and stem elongation, were influenced by the evaluation period (Table 1). Leaf appearance rate directly influences plant morphogenesis, impacting final leaf size, tiller density, and number of live leaves per tiller. The maximum point found for the variable was 34 days, and the estimated response was 0.219 tillers leaf⁻¹ day⁻¹. In general, higher leaf appearance rates are associated with smaller leaves in greater quantity since the faster the appearance of new leaves, the shorter the available time for their full expansion. An upright growth habit, as in Aruana grass, induces an increase in leaf sheath length, which increases the time required for leaf appearance (Negri et al., 2019).

Aerial tiller density revealed an interaction effect ($P = 0.0373$) between the use of nitrogen fertilization and evaluation days (Table 2). Basal tiller density exhibited a linear effect ($P = 0.0001$) with evaluation days (Table 2). The amount of light reaching the plant not only depends on the time span but also directly affects blade growth, as evidenced by the greater development of the leaves in full sun conditions, when the leaves have the potential to achieve greater lengths over shorter evaluation periods.

The reduced leaf lifespan was a response to leaf appearance and elongation

rates (Table 1). The response patterns observed in number of live leaves and leaf lifespan (Table 2) imply a heightened rate of tissue turnover prompted by nitrogen application. This can positively impact the production system as younger leaves exhibit superior photosynthetic capacity. Leaf length is considered an escape characteristic responsive to defoliation intensity, signifying an adaptive characteristic for grazing plants (Costa et al., 2018). In ideal growth conditions, changes in forage tillering are evident, which can be caused by changes in temperature and light regime (Bastos Ongaro et al., 2023).

Aerial and basal tiller density is influenced by nutritional, environmental, and management factors, which collectively shape plant responses to given conditions. The generation of new tillers is dependent on the amount of energy sourced from photosynthesis and the dynamic activity of growth points. This process remains continuous and is expedited when environmental luminosity conditions at the base of the canopy are enhanced (Costa et al., 2018).

The observed decrease in both basal and aerial tiller densities in the silvopastoral system aligns with findings by Paciullo et al. (2017) involving *Panicum maximum* cultivars Tanzania and Massai. These prior studies underscored the significance of sunlight in the broader plant production spectrum. The linear response noted for basal tiller density across evaluation days suggests a strategic emphasis on nurturing preexisting morphogenetic structures, possibly at the expense of generating new ones. This is a common survival-oriented characteristic observed in plants (Costa et al., 2018).

The favorable outcomes concerning the number of leaves per tiller can be attributed to the substantial nitrogen content utilized. This N content contributes significantly to the plant's photosynthetic response to light, enhancing its efficiency in harnessing solar radiation. Genetic characteristics of the forage species play a more dominant role in influencing the number of live leaves per tiller compared to the provided environmental conditions (Paciullo

et al., 2017). Consequently, the impact of N fertilization on number of leaves might be relatively minor. Comparatively lower values have been reported in the literature, such as 3.71 leaves per tiller at the 42-day evaluation mark for cultivar Tanzânia-1 isolated from grazing. This difference could be attributed to its greater height and subsequent reduced tillering in comparison to cv. Aruana (Costa et al., 2018).

Table 1
Effect of nitrogen fertilization and shading on leaf appearance rate, leaf elongation rate, stem elongation rate, leaf senescence rate, leaf lifespan, number of live leaves, final intact leaf length, final defoliated leaf length, aerial tiller density, basal tiller density, leaf area index, total dry matter yield, herbage mass, and daily accumulation rate of Megathyrus maximus

Fertilization (F)	LAR	LER	SER	SLR	LLS	NLL	FILL	FDLL	ATD	BTD	CH	LAI	TDMY	HM	DAR	SD	BWG
Without	0.185	1.197	0.11	0.019	42.22	4.49	9.53	4.98	90.90	533.49	23.59	1.124	13,994.50	1,237.50	45.24	154.55	20.03
With	0.168	1.359	0.114	0.018	40.70	4.33	10.59	5.23	106.89	622.38	25.97	1.231	15,182.00	1,891.30	72.82	145.67	13.26
System (S)																	
Full sun	0.179	1.329	0.117	0.021	42.87	4.56	10.54	5.43	128.47	671.27	21.97	1.604	16,860.00	1,868.69	74.89	166.27	18.96
Silvopastoral	0.173	1.235	0.098	0.017	40.07	4.26	9.60	4.81	71.18	491.20	27.52	0.773	12,316.50	1,259.17	43.16	133.95	14.39
Day																	
21	0.129	1.697	0.328	0.010	51.32	5.46	18.52	8.34	78.67	665.50	32.08	2.052	14,666.00	1,598.33	93.89	132.39	25.67
42	0.213	1.218	0.049	0.045	38.48	4.09	8.56	4.62	87.77	599.11	27.21	1.191	14,033.00	1,692.73	57.62	150.56	12.46
63	0.207	1.054	0.031	0.009	33.30	3.54	5.49	3.20	142.81	575.54	21.57	0.818	15,684.00	1,738.33	52.70	167.38	11.74
84	0.155	1.226	0.045	0.013	43.11	4.59	7.95	4.69	85.33	475.15	17.69	0.673	13,970.00	1,211.67	31.90	-	-
Mean	0.176	1.280	0.108	0.018	41.45	4.40	10.08	5.11	99.01	578.59	24.80	1.178	14,588.25	1,557.45	59.03	150.11	16.74
SEM	0.006	0.051	0.013	0.003	1.119	0.119	0.495	0.213	7.357	18.398	1.213	0.123	1,287.41	97.715	5.881	4.707	1.705
<i>p</i> -value																	
Fertilization	0.2105	0.1186	0.1483	0.7558	0.3617	0.3617	0.0209	0.2054	0.3402	0.0035	0.0318	0.2208	0.0713	0.0001	0.0039	0.2045	0.0011
System	0.6107	0.2784	0.7471	0.2153	0.1198	0.1198	0.2132	0.0508	0.0007	0.0001	0.0002	0.0001	0.0095	0.0001	0.0007	0.0001	0.0158
Day	0.0001	0.0001	0.0001	0.1705	0.0001	0.0001	0.0001	0.0001	0.6318	0.0012	0.0001	0.2633	0.7671	0.2792	0.1942	0.0014	0.0007
F × S	0.8961	0.9984	0.9829	0.5906	0.6341	0.6341	0.4916	0.6089	0.4450	0.2330	0.2791	0.0886	0.0882	0.0221	0.3102	0.4632	0.1732
F × Day	0.1546	0.3936	0.1777	0.4727	0.1284	0.1284	0.8061	0.7419	0.0373	0.9773	0.2072	0.9521	0.4761	0.8240	0.9896	0.9661	0.0223
S × Day	0.9005	0.1740	0.4068	0.4833	0.0195	0.0195	0.9165	0.0168	0.6630	0.6046	0.3610	0.0044	0.2632	0.3583	0.4618	0.8537	0.0334
F × S × Day	0.5380	0.8931	0.1929	0.3538	0.9177	0.9177	0.7262	0.4685	0.0051	0.6401	0.7986	0.3928	0.2317	0.3741	0.0905	0.9246	0.9896

LAR - leaf appearance rate (leaves tiller⁻¹ day⁻¹); LER - leaf elongation rate (mm tiller⁻¹ day⁻¹); SER - stem elongation rate (mm tiller⁻¹ day⁻¹); LSR - leaf senescence rate (mm tiller⁻¹ day⁻¹); LLS - leaf lifespan (days); NLL - number of live leaves (leaves tiller⁻¹); FILL - final intact leaf length (mm); FDLL - final defoliated leaf length (mm); ATD - aerial tiller density (tillers m⁻²); BTD - basal tiller density (tillers m⁻²); CH - canopy height (cm); LAI - leaf area index; TDMY - total dry matter yield (kg ha⁻¹); HM - herbage mass (kg ha⁻¹); DAR - daily accumulation rate (kg ha⁻¹ day⁻¹); SD - stocking density (kg LW ha⁻¹); BWG - body weight gain (kg ha⁻¹). Means followed by different letters in the same column comparing the effect of fertilization and/or shading differ by the T test (p<0.05).

Table 2

Regression equations, coefficients of determination (R²), estimated variable response, minimum or maximum contents (MC), and coefficient of variation (CV%) of *Megathyrus maximus* in silvopastoral and full-sun systems

Variable	Regression equation	R ²	Estimated response	MC	CV
LAR	0.131420 + 0.005168*Day - 0.000076*Day ²	0.246	0.219	34	34.81
LER	1.698065 - 0.003078*Day + 0.000369*Day ²	0.156	1.692	04	42.40
SER	0.315223 - 0.014400*Day + 0.000163*Day ²	0.397	-0.003	44	84.39
BTD	667.8807335 - 2.8208548*Day	0.092	-	-	35.45
FILL	18.450990 - 0.608909*Day + 0.007037*Day ²	0.745	5.279	43	29.23
SD	132.6176389 + 0.8329960*Day	0.262	-	-	16.39
CH	31.96870940 - 0.23251077*Day	0.467	-	-	24.20
Silvopastoral					
NLL	5.840912023 - 0.139870126*Day + 0.001846306*Day ²	0.359	3.192	34	31.16
LLS	54.90457302 - 1.31477918*Day + 0.01735528*Day ²	0.359	30	30	31.16
LAI	1.397390244 - 0.042675184*Day + 0.000451514*Day ²	0.824	0.389	47	25.56
FDLL	8.766770186 - 0.295013803*Day + 0.003528393*Day ²	0.715	2.60	42	30.10
BWG	19.81000000 - 0.72654762*Day + 0.01307823*Day ²	0.239	9.71	27	55.72
Full sun					
NLL	5.177785479 - 0.064142189*Day + 0.000914612*Day ²	0.155	4.053	35	22.94
LLS	48.67118350 - 0.60293658*Day + 0.0085973*Day ²	0.155	38	38	22.94
LAI	2.970666667 - 0.06622619*Day + 0.000519841*Day ²	0.765	0.861	64	28.11
FDLL	7.9596078430 - 0.195939932*Day + 0.002420005*Day ²	0.416	3.993	40	32.29
BWG	31.52666667 - 1.15968254*Day + 0.01604308*Day ²	0.692	10.57	36	34.51

LAR - leaf appearance rate (leaf tiller⁻¹ day⁻¹); LER - leaf elongation rate (mm tiller⁻¹ day⁻¹); SER - stem elongation rate (mm tiller⁻¹ day⁻¹); BTD - basal tiller density (tillers m⁻²); FILL - final intact leaf length (mm); SD - stocking density (kg LW ha⁻¹); CH - canopy height (cm); NLL - number of live leaves (tiller⁻¹ leaves); LLS - leaf lifespan (days); LAI - leaf area index; FDLL - final defoliated leaf length (mm); BWG - body weight gain (kg).

Dry matter yield and animal performance

In terms of dry matter yield and animal performance, nitrogen fertilization exhibited a significant influence (P < 0.05) in full-sun conditions, leading to higher values in herbage mass and daily accumulation rate (Table 1). These findings align with the increased LAI observed (P = < 0.0001) in the full-sun treatment. Notably, herbage mass

production was influenced by the production system, with greater yield (P = < 0.0001) in full sun (Table 1). The higher herbage accumulation rate when combined with nitrogen fertilization and full sun suggests a notable response of plants to the applied fertilizer rate, considering that DM yield and biomass accumulation are impacted in situations with 50% or more sunlight exposure (Barnes et al., 2015).

Stocking density showed a notable increase ($P < 0.0001$) under full-sun conditions (Table 1). The factors influencing animal body weight gain included the evaluation system ($P = 0.0159$), use of N fertilization ($P = 0.0011$), and evaluation period ($P = 0.0007$) (Tables 1 and 2). The full-sun system demonstrated greater favorability for pasture development and production, with a mean surpassing that of the silvopastoral system ($16,860.00 \text{ kg DM ha}^{-1}$). Meeting the carbon C and nitrogen N requirements of the plant triggers meristem activation, resulting in heightened tillering, leaf growth, and biomass accumulation (Assis Farias et al., 2018). The evaluation period was characterized by elevated temperatures, leading to an increased number of leaves and subsequent tillering, largely due to the use of solar luminosity by N, subsequently enhancing leaf crude protein (CP) levels. This, in turn, facilitated augmented herbage production and stocking density.

Chemical composition

The contents of dry matter, CP, neutral detergent fiber, acid detergent fiber, and lignin in leaf and stem samples differed significantly ($P < 0.05$) according to the evaluation day (Table 3). Full sun exposure led to higher CP levels in both leaves and stems (Table 3). This increase in CP content was more pronounced under full-sun conditions, attributable to the influence of sunlight on the plant's utilization of fertilizer and other nutrients. Additionally, the soil-climatic conditions during the experimental phase facilitated a substantial production of leaf blades by the plants, which explains the heightened nutritional content.

Table 3

Dry matter, crude protein, acid detergent fiber, neutral detergent fiber, and lignin contents of samples of leaves and stems of *Megathysus maximus* in silvopastoral and full-sun systems

Fertilization (F)	Leaf					Stem				
	DM	CP	ADF	NDF	LIG	DM	CP	ADF	NDF	LIG
Without	21.93a	18.62b	35.61a	68.73a	2.23	20.36a	10.10b	45.59	76.34a	2.11b
With	21.43b	21.64a	34.91b	66.73b	2.26	19.75b	10.80a	45.37	75.24b	2.50a
System (S)										
Full sun	22.05a	21.81a	34.65b	67.31	2.17b	19.45b	11.02a	44.06b	74.68b	2.23b
Silvopastoral	21.26b	18.45b	35.86a	68.14	2.33a	20.66a	9.88b	46.90a	76.91a	2.39a
Day										
21	20.52	23.71	32.64	62.73	2.26	19.82	11.30	42.09	71.44	2.21
42	23.25	18.80	35.78	68.57	2.37	18.18	10.96	44.17	75.65	2.16
63	18.88	18.48	36.68	70.57	2.19	18.15	8.99	49.18	79.51	2.49
84	24.94	19.52	35.93	69.04	2.14	23.96	10.54	46.47	76.57	2.33
Mean	21.68	20.13	35.26	67.73	2.24	20.06	10.45	45.48	75.79	2.31
SEM	0.447	0.633	0.341	0.689	0.048	0.525	0.305	0.661	0.684	0.069
<i>p</i> -value										
Fertilization	0.0001	<.0001	0.0153	0.0008	0.3891	<.0001	0.0286	0.5171	0.0072	<.0001
System	<.0001	<.0001	0.0003	0.1050	0.0126	<.0001	0.0014	<.0001	<.0001	0.0183
Day	<.0001	<.0001	<.0001	<.0001	0.0170	<.0001	0.0039	<.0001	<.0001	0.0002
F × S	<.0001	0.0751	0.3482	0.9524	0.0716	<.0001	0.5840	0.8493	0.5419	0.0137
F × Day	<.0001	<.0001	0.0429	0.0085	<.0001	<.0001	0.0027	0.0002	<.0001	<.0001
S × Day	<.0001	<.0001	0.9248	0.0005	0.0152	<.0001	0.0004	<.0001	<.0001	<.0001
F × S × Day	<.0001	<.0001	0.0131	0.0084	0.0048	<.0001	0.1019	0.0019	0.0509	0.0004

DM - dry matter (%); CP - crude protein (% DM); ADF - acid detergent fiber (% DM), NDF - neutral detergent fiber (% DM); LIG - lignin (% DM).

Means followed by different letters in the same column comparing the effect of fertilization and/or shading differ by the T test ($p < 0.05$).

The application of nitrogen fertilization at higher rates induces an increase in the fibrous component of the plants (Neumann et al., 2019), especially in older plants, where higher productivity necessitates additional structural support. The higher CP content, coupled with reduced acid detergent fiber and neutral detergent fiber levels, signifies enhanced nutritional value in plants exposed to full sun. This, in turn, influenced herbage

mass production, which was notably higher under sunlight-exposure conditions in conjunction with nitrogen application.

Conclusions

The integration of the silvopastoral system and nitrogen fertilization exhibited positive effects on forage growth, leading to

increased canopy height as well as enhanced rates of leaf and stem appearance and elongation. Moreover, nitrogen fertilization also favored basal tiller density, promoting robust tissue turnover and yielding young and nutrient-rich leaves. Nevertheless, the full-sun system notably excelled in multiple aspects, including dry matter yield, daily accumulation rate, and chemical composition. This was evident in its higher levels of crude protein and decreased fiber content. Consequently, the initial hypothesis proposing that the combination of the silvopastoral system with nitrogen fertilization would enhance forage growth is confirmed. However, it is essential to underscore that the full-sun system demonstrated more favorable outcomes in both herbage yield and quality.

Acknowledgments

This research was carried out with the support of the Coordination for the Improvement of Higher Education Personnel - Brazil (CAPES) - Finance Code 001.

References

- Abraham, E. M., Kyriazopoulos, A. P., Parissi, Z. M., Kostopoulou, P., Karatassiou, M., Anjalanidou, K., & Katsouta, C. (2014). Growth, dry matter production, phenotypic plasticity, and nutritive value of three natural populations of *Dactylis glomerata* L. under various shading treatments. *Agroforestry Systems*, 88(2014), 287-299. doi: 10.1007/s10457-014-9682-9
- Alvares, C. A., Stape, J. L., Sentelhas, P. C., Gonçalves, J. L. M., & Sparovek, G. (2013). Koppen's climate classification map for Brazil. *Meteorologische Zeitschrift*, 22(6), 711-728. doi: 10.1127/0941-2948/2013/0507
- Anjos, A. J., & Chaves, C. S. (2021). Características do componente forrageiro em sistemas silvipastoris. *Scientific Electronic Archives*, 14(3), 53-64. doi: 10.36560/14320211239
- Araújo, S. A. D. C., Silva, T. O. D., Rocha, N. S., & Ortêncio, M. O. (2017). Growing tropical forage legumes in full sun and silvopastoral systems. *Acta Scientiarum. Animal Sciences*, 39(2017), 27-34. doi: 10.4025/actascianimsci.v39i1.32537
- Assis Farias, J. de, F^o., Paula, F. L. M. de, Paula, A. L. de, Paris, W., Ghinzelli, F., Arend, G. H., & Menezes, L. F. G. de. (2018). Production and quality of Tifton 85 pastures overseeded with black oat: effects of irrigation and nitrogen fertilization. *Semina: Ciências Agrárias*, 39(5), 2071-2080. doi: 10.5433/1679-0359.2018v39n5p2071
- Association of Official Analytical Chemists (2016). Official methods of analysis of AOAC International. In G. W. Latimer Jr. (Ed.), Official methods of analysis of AOAC International (20nd ed., pp. 3172). Rockville, Maryland, USA: AOAC.
- Barnes, P., Wilson, B. R., Reid, N., Bayerlein, L., Koen, T. B., & Olupot, G. (2015). Examining the impact of shade on above-ground biomass and normalized difference vegetation index of C3 and C4 grass species in North-Western NSW, Australia. *Grass and Forage Science*, 70(2), 324-334. doi: 10.1111/gfs.12118

- Bastos Ongaro, A. F., Azevedo Martuscello, J., Andrade Gimenes, F. M. de, Lopes Batista, A. C., Ferreira Penteadó, L., Premazzi, L. M., Mattos, W. T., Vieira, M. M., Costa, R. L. D. da, & Gerdes, L. (2023). Canopy height impact on legume mass and *Megathyrsus maximus* tiller dynamics in mixed pastures. *Acta Agriculturae Scandinavica*, 73(1), 114-126. doi: 10.1080/09064710.2023.2226650
- Carrère, P., Louault, F., & Soussana, J. F. (1997). Tissue turnover within grass-clover mixed swards grazed by sheep fluxes. *Journal of Applied Ecology*, 34(1997), 333-348. doi: 10.2307/2404880
- Cipriani, H. N., Salman, A. K. D., Passos, A. M. A. dos, Schmitt, E., Cruz, P. C., Botelho, F. J. E., & Moraes, K. K. S. (2016). *Uma planilha eletrônica gratuita para calcular a sombra projetada pelas árvores*. (Circular técnica, 145). EMBRAPA. <https://ainfo.cnptia.embrapa.br/digital/bitstream/item/153440/1/CT-145-Planilha-de-Sombra.pdf>
- Costa, N. D. L., Jank, L., Magalhães, J. A., Rodrigues, A. N. A., Fogaça, F. D. S., Bendahan, A. B., & Santos, F. D. S. (2018). Características morfogênicas e estruturais de *Megathyrsus maximus* cv. Tanzânia-1 sob intensidades de desfolhação. *PUBVET*, 12(4), 1-7. doi: 10.22256/pubvet.v12n4a67.1-7
- Empresa Brasileira de Pesquisa Agropecuária (1999). *Sistema brasileiro de classificação dos solos*. EMBRAPA Solos.
- Fagan, E. B., Ono, E. O., Rodrigues, J. D., Soares, L. H., & Dourado, D., Neto. (2016). *Fisiologia vegetal: metabolismo e nutrição mineral*. Editora Andrei.
- Grupo de Pesquisa em Ciência do solo - GPCS. (2022, Setembro, 10). *Dados clima DV 2019; 2020*. <https://www.gebiomet.com.br/pt/downloads>
- Klingman, D. L., Miles, S. R., & Mott, G. O. (1943). The cage method for determining consumption and yield of pasture herbage. *Journal of the American Society Agronomy*, 35(9), 739-746. doi: 10.2134/agronj1943.00021962003500090001x
- Komareck, A. R. (1993). A filter bag procedure for improved efficiency of fiber analysis. *Journal of Dairy Science*, 76(Suppl. 1), 250-259.
- Lemaire, G., & Chapman, D. (1996). Tissue flows in grazed plant communities. In J. Hodgson, & A. W. Illus, A. W. (Eds.), *Morphogenesis and tiller density of Aruana grass managed at different heights under sheep grazing* (pp. 3-36). Amsterdam.
- Lima Veras, E. L. de, Difante, G. D. S., Chaves Gurgel, A. L., Graciano da Costa, A. B., Gomes Rodrigues, J., Marques Costa, C., Emerenciano, J. V., Neto, Pereira, M. G., & Ramon Costa, P. (2020). Tillering and structural characteristics of *Panicum* cultivars in the Brazilian semiarid region. *Sustainability*, 12(9), 3849. doi: 10.3390/su12093849
- López-Carrasco, C., López-Sánchez, A., San Miguel, A., & Roig, S. (2015). The effect of tree cover on the biomass and diversity of the herbaceous layer in a Mediterranean dehesa. *Grass and Forage Science*, 70(4), 639-650. doi: 10.1111/gfs.12161
- Mott, G. O., & Lucas, H. L. (1952). The design, conduct, and interpretation of grazing trials on cultivated and improved pastures.

- Proceeding of the International Grassland Congress, State College, Proceedings State College, Pasadena, Pennsylvania, EUA.*
- Neel, J. P. S., Felton, E. E. D., Singh, S., Sextone, A. J., & Belesky, D. P. (2016). Open pasture, silvopasture and sward herbage maturity effects on nutritive value and fermentation characteristics of cool-season pasture. *Grass and Forage Science*, 71(2), 259-269. doi: 10.1111/gfs.12172
- Negri, R., Santos, G. B. dos, Paulo Macedo, V. de, Silveira, M. F. da, Wlodarski, L., & Kluska, S. (2019). Morphogenesis and tiller density of Aruana grass managed at different heights under sheep grazing. *Semina: Ciências Agrárias*, 40(5 Suppl. 1), 2341-2350. doi: 10.5433/1679-0359.2019v40n5Supl1p2341
- Neumann, M., Horst, E. H., Souza, A. M. de, Venancio, B. J., Stadler, E. S., Jr., & Karpinski, R. A. K. (2019). Avaliação de doses crescentes de nitrogênio em cobertura em milho para silagem. *Agrarian*, 12(44), 156-164. doi: 10.30612/agrarian.v12i44.7195
- Paciullo, D. S. C., Gomide, C. D. M., Castro, C. D., Maurício, R. M., Fernandes, P. B., & Morenz, M. J. F. (2017). Morphogenesis, biomass and nutritive value of Panicum maximum under different shade levels and fertilizer nitrogen rates. *Grass and forage Science*, 72(3), 590-600. doi: 10.1111/gfs.12264
- Paciullo, D. S. C., Pires, M. F. A., Aroeira, L. J. M., Morenz, M. J. F., Maurício, R. M., Gomide, C. A. M., & Silveira, S. R. (2014). Sward characteristics and performance of dairy cows in organic grass-legume pastures shaded by tropical trees. *Animal*, 8(8), 1264-1271. doi: 10.1017/S1751731114000767
- Sacramento, A. M. H., Menezes, O. C. de, Barros, T. M., Pinheiro, D. N., Jaeger, S. M. P. L., Ribeiro, O. L., Ramos, C. E. C. O., & Oliveira, G. A. de. (2019). Morphogenic and structural characteristics and chemical composition of grass aruana, submitted to nitrogen fertilization. *Semina: Ciências Agrárias*, 40(6 Suppl. 2), 3167-3180. doi:10.5433/1679-0359.2019v40n6Supl2p3167
- Santiago-Hernández, F., López-Ortiz, S., Ávila-Reséndiz, C., Jarillo-Rodríguez, J., Pérez-Hernández, P., & Guerrero-Rodríguez, J. D. (2016). Physiological and production responses of four grasses from the genera *Urochloa* and *Megathyrsus* to shade from *Melia azedarach* L. *Agroforestry Systems*, 90(2016), 339-349. doi: 10.1007/s10457-015-9858-y
- Statistical Analysis System (2017). *Sas/Stat University User Guide*. Sas Institute Inc.
- Senger, C. C. D., Kozloski, G. V., Sanchez, L. M. B., Mesquita, F. R., Alves, T. P., & Castagnino, D. S. (2008). Evaluation of autoclave procedures for fibre analysis in forage and concentrate feedstuffs. *Animal Feed Science and Technology*, 146(1-2), 169-174. doi: 10.1016/j.anifeedsci.2007.12.008
- Silveira, M. L., Vendramini, J. M. B., Sellers, B., Monteiro, F. A., Artur, A. G., & Dupas, E. (2015). Bahiagrass response and N loss from selected N fertilizer sources. *Grass and Forage Science*, 70(1), 154-160. doi: 10.1111/gfs.12078

- Sousa, L. F., Santos, J. G. D., Alexandrino, E., Maurício, R. M., Martins, A. D., & Sousa, J. T. L. (2015). Método prático e eficiente para estimar a área foliar de gramíneas forrageiras tropicais. *Archivos de Zootecnia*, 64(245), 83-85. doi: 10.21071/az.v64i245.380
- Van Soest, P. J., Robertson, J. B., & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. *Journal of Dairy Science*, 74(10), 3583-3597. doi: 10.3168/jds.S0022-0302(91)78551-2
- Wilm, H. G., Costello, D. F., & Klipple, G. E. (1944). Estimating forage yield by the 11 double-sampling methods. *Journal of American Society of Agronomy*, 36(3), 194-203. doi: 10.2134/agronj1944.00021962003600030003x
- Zanchi, F. B., Aguiar, L. J., Von Randow, C., Kruijt, B., Cardoso, F. L., & Manzi, A. O. (2009). Estimativa do índice de área foliar (IAF) e biomassa em pastagem no estado de Rondônia, Brasil. *Acta Amazonica*, 39(2009), 335-347. doi: 10.1590/S0044-59672009000200012

