BIOSCIENCE JOURNAL

ANATOMICAL, MORPHOGENIC AND STRUCTURAL CHARACTERISTICS OF XARAÉS PALISADE GRASS UNDER GRAZING

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How to cite: BASSO, K.C., et al. Anatomical, morphogenic and structural characteristics of Xaraés palisade grass under grazing. *Bioscience Journal*. 2023, **39**, e39067. https://doi.org/10.14393/BJ-v39n0a2023-60937

Abstract

This study evaluated under grazing intensities and periods of the year: leaf anatomy of *Urochloa brizantha* cv. Xaraés and its correlation with morphogenetic and structural characteristics, and leaves degradation after in situ incubation. Treatments were four grazing intensities (GI) defined by the pasture residuals leaf area index (rLAI 0.8, 1.3, 1.8, and 2.3) with three replications in a completely randomized design. Cows grazed in a rotational stocking with pastures regrowth period determined by 95% light interception. Leaves showed a higher proportion of sclerenchyma (2.64%) in pastures under lower GI and in the dry season (2.42%). Pastures managed under higher GI showed lower number of expanded leaves (2.58), lower number of lives leaves (3.45), and lower leaf senescence rate (0.05 cm tiller−1 d−1). Positive correlation was observed between leaf elongation rate and adaxial epidermis and vascular tissues. rLAI 1.8 and 2.3 provided greater residues after in situ leaf incubation at times 12, 48, 72, and 96 h compared to rLAI 0.8 and 1.3. rLAI and period of the year had little influence on leaf anatomy of the Xaraés managed under 95% LI, and leaf anatomy is correlated with the morphogenetic and structural pasture characteristics. Pastures managed under lower GI show more residues after leaves incubation in rumen.

Keywords: Light interception. Microscopy. Rumen incubation. *Urochloa.*

1. Introduction

Plants can adapt to various climate changes and type of management imposed. This adaptation occurs mainly due to its phenotypic plasticity, with complex adjustments in plant physiology and in the structure of leaf tissues (Habermann et al. 2019). Changes in the structure of leaf tissues in grasses, in turn, can cause changes in the chemical composition of forage and thus can interfere in the forage intake and animal performance (Moore et al. 2020).

To better understand leaf tissue changes in grasses, studies such as leaf anatomy associated with morphogenic and structural evaluations can be very important, because with these studies we know the proportion of tissues according to species, vegetative stage, seasons of the years, and in function of different managements. With these associated studies we can understand a little more about forage quality, once the chemical composition, digestibility and canopy structure do not always explain all variations in forage intake.

Studies about physiology and morphology of tropical grasses are important (Maranhão et al. 2021; Mussso et al. 2021; Solofondranohatra et al. 2021) mainly when related to variations in leaf anatomy and functional responses of forage plants to grazing. Thus, these studies help us to identify and strategically plan the grazing management, providing longevity, productivity, and sustainability to the pasture ecosystems. In this sense, in a study carried out by Mauri et al. 2018, the authors evaluated *Urochloa brizantha*, *Urochloa decumbens, Urochloa ruziziensis* and three clones of *Urochloa ruziziensis* as a function of three regrowth periods (8, 15 and 29 days), and concluded based on the leaf anatomy of the species, that the leaf development of Brachiaria is not complete at 8 days of regrowth. The regrowth age modified the percentage of all tissues in Brachiaria leaves, and fibers, xylem and vascular bundles increased in proportion in older leaves and showed significantly higher proportion at 29 regrowth days.

Coupled with information on the leaf anatomy and morphogenetic characteristics, studies involving light interception by the sward and residual leaf area index as predictors of frequency and grazing intensity respectively, have demonstrated the importance of these variables regard to the growth of plants and responses to defoliation. Regrowth ability, in addition help to define the best moment for the insertion and removal of animals into and from pastures under intermittent grazing (Galzerano et al. 2013; Galzerano et al. 2015; Silva et al. 2016a; Silva et al. 2016b; Silva et al. 2019). Thus, we need to understand how the grazing intensity in intermittent grazing can change the morphogenesis and leaf anatomy of grasses in order to interfere in the degradability of tissues in the animals' rumen.

We attempted to answer the following questions with this study: A) Does grazing intensities, grazing cycles and periods of the year alter leaf anatomy and morphogenetic and structural characteristics? B) Is there any correlation among leaf anatomy and morphogenetic characteristics? C) Could grazing intensities and periods of the year change leaf blade degradability in the rumen? Since grazing intensities and successive grazing cycles promote changes in leaf anatomy, morphogenic and structural characteristics, in addition to the period of the year, we hypothesized that lower grazing intensities and the dry period could increase leaf senescence, lifespan leaf, and more lignified tissues (vascular and parenchymal tissues) and these facts will result in lower leaf blade degradability.

2. Material and Methods

The study was conducted from January to June 2010 and split in two periods (rainy and dry) on Xaraés palisadegrass (*Urochloa brizantha* cv. Xaraés) pastures seeded since 2004 in Jaboticabal, SP, Brazil (21º15’S, 48º18’W). The local climate is characterized as an Aw type, according to the Köppen classification, with two distinct periods: dry, from April to September; and rainy, from October to March. Total precipitation during the collection period was 1,329 mm, and the average temperature was 23.6 ºC.

The experimental area is located at 595 m above the sea level and the soil is a Rodhic Ferralsol (IUSS, 2014). The following properties were revealed in the analysis performed in the 0-20 cm layer: 27 mmol dm^{-3} Ca^{2+}; 9.5 mmol dm^{-3} Mg^{2+}; 3.1 mmoldm^{-3} K^+; 10.0 mg dm^{-3} P (resin); 22.5 g dm^{-3} organic matter; and 4.9 pH ($CaCl_2$).

The total Xaraés palisadegrass area was 2795.9 m^2, divided into 12 paddocks for allocating the treatments. Treatments consisted of four grazing intensities, defined by four residual leaf area index of the pastures (rLAI of 0.8, 1.3, 1.8, and 2.3), arranged in a completely randomized design with three replications. Two months before the start of the evaluations (November 2009) grazing was started to ensure that the desired rLAI were achieved. The mob-stocking technique was adopted for grazing, using groups of animals for rapid defoliations, simulating an intermittent grazing scenario (Allen et al. 2011). For this purpose, non-lactating Holstein cows (Bos taurus taurus L.) with an approximate 450 kg BW were used.

The rLAI values were chosen based on previous experiments conducted by the research group in pastures of Tifton 85 (Cynodon spp.) (Silva et al. 2013; Costa et al. 2015; Silva et al. 2016a; Silva et al. 2019) and Xaraés palisadegrass (Raposo et al. 2014; Galzerano et al. 2015) in order to cover an amplitude that represented different grazing intensities. The rLAI was monitored during the grazing period through measurements taken in each paddock, and once the rLAI was achieved, animals were moved to another paddock where the pasture had intercepted 95% of light (LI) during the regrowth period. A total of four grazing cycles were carried out during the entire experimental period, each defined as the sum of the paddock occupation period and the pasture regrowth period. In the rainy period (January to April) three grazing cycles were carried out, and in the dry period (May and June) only one grazing cycle was carried out.

Light interception (LI) was monitored from the moment of the animals were removed from the paddocks and during the pasture regrowth period until they reached 95% LI. During the pasture regrowth period, LI was read weekly, until it was near 93%, which was when readings were taken daily until 95% LI, moment when the LAI was recorded (LAI in the pre-grazing, denominated preLAI). Twenty readings were performed for LI, preLAI and rLAI per paddock, always between 11h00 and 13h00, using an AccuPAR LP-80 ceptometer (Decagon Devices, Pullman, WA).

To determine the percentage of the grass leaf tissues, collections were performed when the pastures reached 95% LI (pre-grazing), with nine leaves collected from three vegetative tillers (three leaves per tiller) in each paddock in January, February, and April (rainy period), and May and June (dry period). The last leaves with exposed ligules were selected, sectioned at the collar, and packed in plastic bags. Immediately after being harvested, leaves were cut in the medial region, generating fragments with approximately 1 cm that were stored in 10 mL glasses and covered with a 50% formalin-acetic acid-alcohol solution until the beginning of histological preparations.

Fragments of each leaf were subjected to an alcoholic series with tertiary butyl alcohol for approximately 40 h to gradually remove the water and avoid cell plasmolysis and progressively dry the material, as described by Basso et al. (2014). After the alcoholic series, fragments were paraplast embedded after being sectioned transversally at 10 µm using a rotary microtome. Subsequently samples were deparaffinized and tissues were stained with Triarch's Quadruple Stain. Permanent slides were mounted for anatomical quantification.

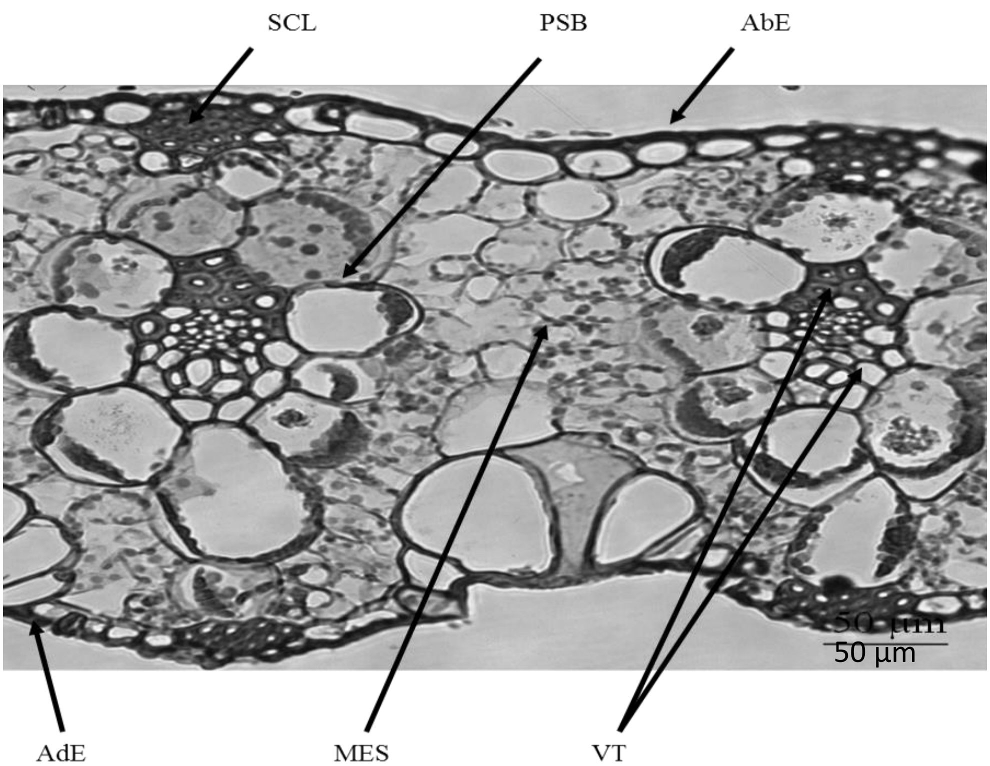

Figure 1. Cross-section of a Xaraés palisadegrass leaf blade. Note. SCL=sclerenchyma, PSB=parenchyma sheath of vascular bundles, AbE=abaxial epidermis, AdE=adaxial epidermis, MES=mesophyll and VT=vascular tissues (50 µm).

The proportions of the different leaf tissues were determined using a binocular optimal microscope coupled to the image analysis software (Axion Vision, version 3.1.). The total area of the cross-section of the leaf blade projected on the video were measured, and then the areas of the adaxial and abaxial epidermis (AdE and AbE), EPI (AdE + AbE), sclerenchyma (SCL), parenchyma sheath of vascular bundles (PSB), and vascular tissues (VT) were measured. Mesophyll (MES) area was calculated as the difference between the total cross-section area and the areas of the other tissues. With the respective tissue's areas, the proportions were calculated. Figure 1 shows the leaf cross-sections indicating the evaluated tissues.

To evaluate the pasture morphogenetic and structural characteristics, four tillers were identified per pasture in different points representative of the average canopy height. Tillers were evaluated every three days, during the entire regrowth period in each grazing cycle. After the animals grazed, new tillers were marked for the evaluation of the following grazing cycle. Stem and leaf lengths were measured, and newly emerged leaves per tiller and expanded and senescent leaves were recorded to determine the following morphogenetic and structural variables: final leaf length (FLL, cm); number of expanded leaves (NEL); number of leaves under expansion (NLUE); number of live leaves (NLL); phyllochron (PHY, days leaf−1 tiller−1); leaf elongation rate (LER, cm tiller−1 day−1); leaf senescence rate (LSR, cm tiller−1 day−1); and leaf lifespan (LLS, days), as described by Galzerano et al. (2013).

For the evaluation of the digestion residue in situ of leaf blade fragments, we randomly collected 20 leaves from each paddock at the moment the pastures reached 95% LI. Two collections were performed — one in the rainy period (January) and another in the dry period (June). Leaf samples were partitioned and incubated in situ using rumen-cannulated cattle. Samples were placed in previously dried, 14 × 7 cm (100 µm) non-woven textile bags of known weight. The equivalent amount of dry matter was four grams, which corresponds to 20 mg cm−2. Three bags were incubated according to incubation time (0, 12, 24, 48, 72, 96, and 240 h) and per treatment. For this procedure, two rumen-cannulated Nellore cattle kept grazing in an area adjacent to the experiment were used.

After each incubation period, bags were removed from the rumen and frozen, and only after this step was over, incubated samples were thawed and washed in running water. Next, bags were dried in a forced-air oven at 65 ºC for 72 h and weighed. The percentage disappearance of the soluble fraction based on the original material was calculated as the initial weight minus the value obtained after oven-drying.

We chose to evaluate the leaf fragments after incubating the samples of pastures under higher grazing intensity (rLAI = 0.8 and 1.3) in comparison with those of pastures under lower grazing intensities (rLAI = 1.8 and 2.3) because of the large number of samples that were incubated, and, especially, differences that were observed only between pastures with higher and lower grazing intensity.

The data were analyzed using the MIXED procedure of SAS (Statistical Analysis System, version 9.2). The rLAI, cycle grazing, period (rainy or dry), and interactions were adopted as fixed effects, and the blocks (replicates) and their interactions were considered random effects. The repeated measures procedure was used for variables with measurements having consecutive cycles (anatomy and morphogenetic and structural variables) using compound symmetry as the covariance structure, due to the lower Akaike value. For the leaf blade incubation, repeated measurement was not used, because only two evaluations were performed, one in the rainy period and the other in the dry period. Adjusted means were compared by Tukey's test, except for the means of morphogenetic and structural characteristics that were evaluated by polynomial orthogonal contrasts. All comparisons were performed at P<0.05. The correlation analyses were performed between the leaf-anatomy and morphogenetic and structural variables, as well as within the anatomical characteristics using the Pearson correlation coefficients and t-test, at P<0.05.

3. Results

In the proportions of leaf tissues, there was no interaction among rLAI, grazing cycle, and period (P≥0.05), being observed isolated effect of rLAI and period (P<0.05) (Table 1). The rLAI affected only the percentage of SCL, which was 19% greater in the leaves from pastures managed under a lesser grazing intensity (rLAI = 2.3) compared to pastures under greater grazing intensity (rLAI = 0.8 and 1.3). The proportion of SCL differed between the rainy and dry period (P<0.05), and the other anatomical characteristics were similar (P≥0.05). The forage harvested at dry period provided a higher percentage of SCL in the leaves of the Xaraés palisadegrass (2.42%) as compared with the rainy period (2.01%).

The morphogenetic and structural variables were not significantly affected by any interaction (P≥0.05) but were affected by rLAI (P<0.05) (Table 2). The variables NEL and NLL increased 22% and 16%

respectively, from rLAI 0.8 to 1.3, and then decreased under rLAI 1.8 and 2.3, which provided the quadratic effect. The LER showed quadratic effect with higher value (3.03 cm tiller^{-1} day^{-1}) under rLAI 1.8 and lower value (2.26 cm tiller^{-1} day^{-1}) under rLAI 2.3 (decrease of 25%). LSR and LLS increased linearly from rLAI 0.8 to rLAI 2.3 (P<0.05), with LSR and LLS increasing 420% and 26% respectively, from the lower to higher rLAI. The preLAI increased linearly from LAI 0.8 to 2.3, which represents an increase of 21%.

A positive correlation was observed between LAI in the pre-grazing with AbE and with VT (P<0.05). Leaf elongation rate (LER) was also positively associated with AbE and with VT (P<0.05) (Table 3).

The percentage of AdE correlated linearly and positively with AbE (P<0.01) and EPI (P<0.001), while the correlation between AdE and PSB and AdE and MES was negative (Table 4). Other linear and positive correlations were observed, such as between AbE and EPI (P<0.001), and between PSB and VT (P<0.05). Linear and negative correlations were observed between AbE and MES (P<0.01), EPI and PSB (P<0.001), EPI and MES (P<0.01), PSB and MES (P<0.001) and between VT and MES (P<0.00.1).

Table 1. Proportion of tissues in the cross-section of leaf blade of Xaraés palisadegrass under different residual leaf area index and periods.

rLAI	AdE	AbE	PSB	VT	SCL	MES
			Proportion (%)			
0.8	14.89	5.70	41.30	5.31	2.10 B	30.61
1.3	13.52	4.90	42.60	6.10	2.10 B	30.50
1.8	14.63	6.20	41.61	6.12	1.92 B	29.33
2.3	15.51	5.62	42.53	6.01	2.64 A	27.64
SE	2.31	0.98	3.47	0.71	0.20	1.30
Season						
Rainy	14.30	5.60	41.72	6.00	2.01 B	30.22
Dry	14.90	5.60	42.39	5.80	2.42 A	28.85
SE	1.41	0.61	3.45	0.98	0.25	0.31

Note. AdE=adaxial epidermis; AbE=abaxial epidermis; PSB=parenchyma sheath of vascular bundles; VT=vascular tissue; SCL=sclerenchyma; MES=mesophyll. Means within columns followed by different letters are different by Tukey test at rLAI and by F test at season (P<0.05).

Table 2. Morphogenetic and structural characteristics of Xaraés palisadegrass as a function of rLAI.

rLAI	FLL	NEL	NLUE	NLL	PHY	LER	LSR	LLS	preLAI
0.8	16.50	2.58	0.88	3.45	12.93	2.68	0.05	39.97	4.17
1.3	18.78	3.15	0.87	4.02	13.54	2.79	0.08	37.10	4.48
1.8	19.47	2.73	0.89	3.62	11.19	3.03	0.13	49.52	4.49
2.3	18.96	2.63	0.83	3.67	14.21	2.26	0.26	50.44	5.05
SE	1.77	0.08	0.02	0.07	0.60	0.20	0.04	1.73	0.13
P value	0.6108	0.0002	0.4223	<0.0001	0.1133	0.0047	<0.0001	<0.0001	<0.0001
Contrast†	ns	Q	ns	Q	ns	Q	L	L	L

Note. Final leaf length (FLL, cm); number of expanded leaves (NEL); number of expanding leaves (NLUE); number of live leaves (NLL); phyllochron (PHY, days leaf−1 tiller−1); leaf elongation rate (LER, cm tiller−1 day−1); leaf senescence rate (LSR, cm tiller−1 day−1); leaf lifespan (LLS, days) and pre-grazing leaf area index (preLAI). †Contrast: ns=not significant; L=linear and Q=quadratic.

Table 3. Linear correlation coefficient between morphogenic and structural characteristics and leaf anatomical variables of Xaraés palisadegrass.

Variables	AdE	AbE	EPI	PSB	VT	SCL	MES
preLAI	0.10	0.34**	0.21	-0.12	0.46***	-0.20	-0.22
FLL	-0.01	0.25	0.08	0.06	0.17	0.09	-0.20
NEL	-0.01	-0.20	-0.08	0.23	-0.05	-0.13	-0.07
NLUE	-0.05	-0.11	-0.08	-0.02	-0.17	-0.02	0.12
NLL	-0.02	-0.21	-0.10	0.24	-0.08	-0.14	-0.05
PHY	0.02	-0.23	-0.07	-0.07	0.23	0.27	0.16
LER	0.07	0.28*	0.16	-0.05	0.33**	-0.14	0.21
LSR	0.06	0.16	0.11	-0.09	-0.06	-0.11	0.02
LLS	-0.03	-0.35	-0.16	0.14	-0.28	0.15	0.10

No interaction effect between rLAI and period of the year was observed (P≥0.05) for the residue of digestion in situ of leaf blade, only rLAI effect was observed (P<0.05) (Figure 2). The reduction of residue after in situ digestion was 91.5% from time 0 to 240h. The higher rLAI (1.8 and 2.3) provided higher residues after incubation at times 12, 48 and 96h compared to the lower rLAI (0.8 and 1.3).

Table 4. Linear correlation coefficient between leaf anatomical variables of Xaraés palisadegrass.

Variables	AdE	AbE	EPI	PSB	VT	SCL	MES
AdE	1.00	0.30**	0.93***	-0.50***	-0.09	0.12	-0.28*
AbE		1.00	0.63***	-0.17	0.01	0.36	-0.34**
EPI			1.00	-0.48***	-0.07	0.12	-0.36**
PSB				1.00	0.37*	-0.08	-0.57***
VT					1.00	-0.17	-0.64***
SCL						1.00	-0.08
MES							1.00

Note. AdE=adaxial epidermis; AbE=abaxial epidermis; EPI=epidermis; PSB=parenchyma sheath of vascular bundles; VT=vascular tissue; SCL=sclerenchyma; MES=mesophyll. ***P<0.001, **P<0.01, *P<0.05.

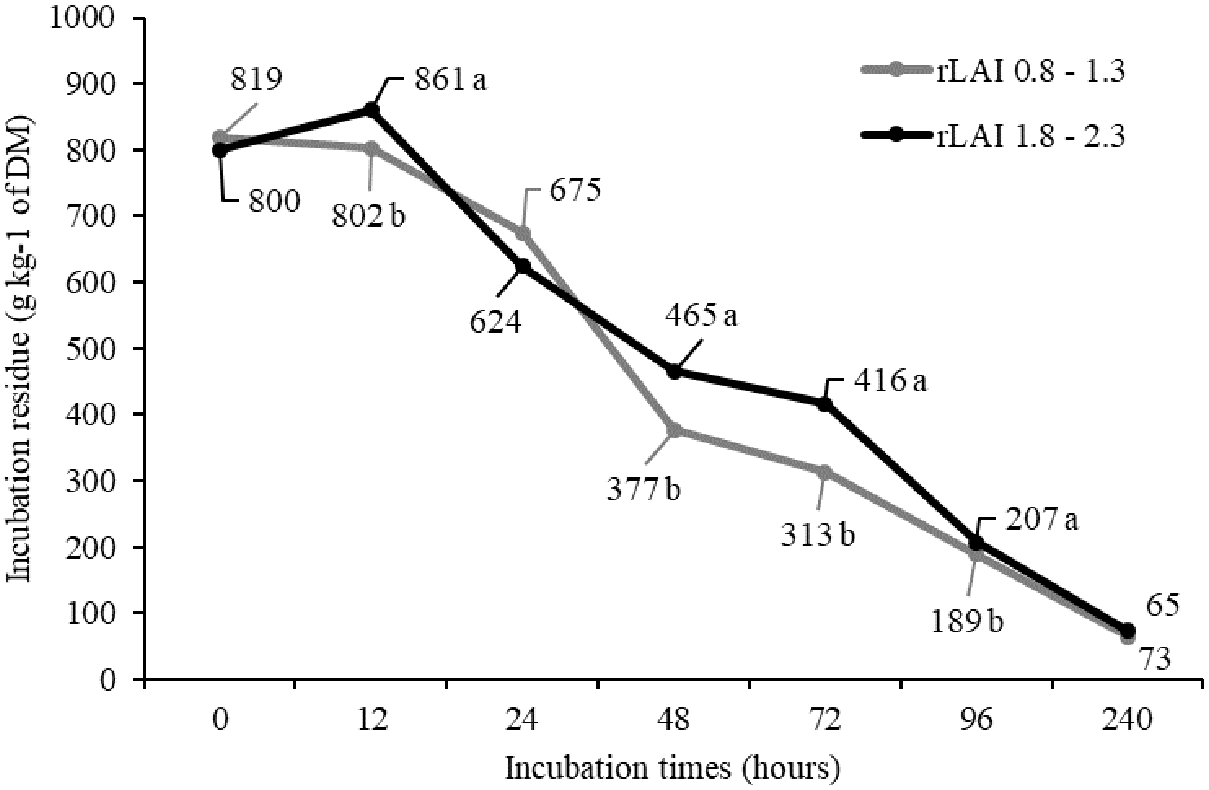

Figure 2. Residue of digestion in situ of leaf blade fragments of Xaraés palisadegrass under different residual leaf area index (rLAI). Means followed by different letters are different by the Tukey test comparing rLAI (P<0.05).

4. Discussion

The average values, in percentage, of AdE and AbE were 14.64 and 5.61, respectively (Table 1). These values are similar to those observed by Mauri et al. (2018) in a study conducted with six *Brachiaria* genotypes. The authors observed average values ranging from 14 to 17% for AdE and from 7.2 to 10% for AbE due to different regrowth ages.

The management of pastures at a lower grazing intensity provided stem elongation and consequently an increase in the stem proportion as well as accumulation of older ungrazed leaves (Euclides et al. 2019) due to the higher rLAI used, which might have increased the proportion of sclerenchyma (Table 1). Studies conducted by Galzerano et al. (2015) demonstrated alterations in leaf:stem ratio and in the proportion of dead material in Xaraés palisadegrass pastures managed under different grazing intensities. Galzerano et al. (2013) also evaluated the effects of rLAI on the morphogenetic traits of Xaraés palisadegrass under grazing and determined a quadratic effect of rLAI on the final leaf length in one of the grazing cycles. These facts suggest that longer leaves may accumulate a larger amount of SCL to support their larger weight and size.

The sclerenchyma may directly or indirectly interfere with the degradability of leaf blades (Sanchês et al. 2018). Directly, depending on how much it represents along the cross-section, because of its indigestible nature. Indirectly the sclerenchyma interferes with degradability depending on its location in the leaf blade, given that it may form a I-girder or T-girder structure. This structure presents an “I” shape, which connects the two sides of the leaf's epidermis, or present a “T” shape, where only one side of the epidermis has this connection (Bauer et al. 2008). The girder structure prevents or make it dificultt to remove the epidermis by digestion and reduces the access of ruminal microorganisms to the mesophyll and parenchyma.

In relation to the constant proportion of the tissues in the cross-section of leaf blade in the rainy and dry season by treatments (Table 1) except for SCL, Habermann et al. (2019) reported that the effects of temperature on the proportion of tissues and leaves of *Panicum maximum* are less obvious than the concentration of atmospheric CO_2. For this reason, decreases in forage digestibility resulting from climatic alterations and the advance in development should not be attributed to alterations in the proportion of leaf tissues, but rather to the intense cell wall lignification.

The increased proportion of SCL in leaf blades during the dry season can be attributed to alterations suffered by parenchymal cells during development. In the dry season, part of the parenchymal cells might have undergone a progressive thickening and lignification of the wall, forming a rigid ring of sclerenchymatous cells.

The observations of no change in the FLL, NLUE and PHY variables can be justified by the criterion used for the beginning of grazing, which was the same for all treatments, ie, 95% light intercept which does not characterize a condition of intense competition for light. However, other variables such as NEL, NLL, LER, LSR and LLS were influenced, demonstrating that for these variables the effects of grazing intensities were more accentuated.

The NLL (Table 2) of grasses is genetically determined, assuming a relatively constant value after the pasture reaches an equilibrium condition, in which the processes of leaf appearance and death are synchronized and may vary with the pasture management and environment conditions (Lemaire and Chapman, 1996; Zanine et al. 2016). Therefore, we can infer that the number of live leaves (NLL) is determined by the sum of expanding (NEL) and expanded (NLUE) leaves per tiller, excluding leaves that present more than 50% of their length already senescent (Montagner et al. 2012). Since the NLUE was not modified by rLAI, the NEL promoted the effects observed in the NLL. Thus, the rLAI 1.3 provided the higher NEL (3.15 leaves) and consequently the higher NLL (4.02 leaves). The pastures managed under rLAI of 2.3 showed higher residue height which may have contributed to the higher LSR, decreasing up both NEL and NLL. In pastures under rLAI of 0.8, due to more intense grazing there are greater chances of apical meristm removal in the tillers, which can stimulate emergence of new tillers with lower number of leaves with reduced length (Barbosa et al. 2021).

In pastures managed up to rLAI 1.8 the LER increased due to higher stem elongation, which provide longer time for leaf emergence resulting in higher LER (Silva et al. 2013). This response pattern is common in tropical grasses when managed under different grazing intensities, i.e., grazing heights (Sbrissia et al. 2018; Oliveira et al. 2020) or rLAI (Silva et al. 2016b). Another justification for this response pattern is due to the lower removal of expanding leaves and stem during grazing in pastures under lower grazing intensities.

The linear increase in LSR with increasing pasture rLAI (Table 2) is due to increased pasture height, which accentuates the shading caused by the upper canopy layers over the lower layers. Silva et al. (2016b) also observed the same response pattern for LSR in Tifton 85 pastures managed under rLAI 0.8, 1.6 and 2.4, with values ranging from 0.33 to 0.46 cm tiller^{-1} day^{-1}. The LLS ranged from 37.10 to 50.44 days with increasing pasture rLAI suggesting greater tissue renewal under more intensive grazing conditions.

The correlation between preLAI and VT (Table 3) can be associated with leaf maturation with increased preLAI of pastures (Table 2). Wilson et al. (1983) found that *Cenchrus ciliaris* genotypes with heavier leaves and a high specific leaf area (SLA) in g DM cm^{-2} were associated with higher percentages of tissues of thick wall (PSB+VT+SCL) in the leaf cross-section. The epidermal and vascular-tissue cells increased their proportions in the leaf as it was elongated because during the period of leaf elongation or expansion, many of the primarily formed tracheal elements of the young xylem (protoxylem) have their secondary walls deposited, which causes their thickening, thereby allowing the tracheids to be distended or extended during the elongation of the organ. Wilson and Hatfield (1997) also reported an increase area of PSB, epidermal, and VT cells with the increase in leaf area and specific leaf area.

The positive correlations between the adaxial and abaxial epidermis (Table 4) are understandable since they are the same tissue, and both differ only in their location in the leaf blade (Figure 1). The positive correlations between AdE and AbE with EPI are due to the EPI originates from the sum of Ade and Abe, so any increase in one of the epidermis will promote an increase in EPI. The epidermis is a leaf tissue directly related to environmental factors because it is in direct contact with the environment. In this sense, in environmental conditions that promote thickening, such as high luminosity (Dickison, 2000), there is probably a reduction in the proportion of other tissues, such as PSB. In this example of high luminosity, the grass normally increases the thickness of the epidermis, and probably does not need excessive elongation of the tillers, which would not justify the elongation and thickening of the PSB.

The positive correlation between PSB and VT (Table 4) can be explained by the increased proportion of PSB, which promotes the increase in VT and SCL, since sheath cells are arranged around VT, and in sclerenchyma is present in each arrangement of these cells, which may or may not form an I- or T-girder structure in C4 grasses.

Mesophyll (MES) was negatively correlated with all the other proportions of leaf tissues, except for SCL (Table 4). This relationship is a result of the calculation used to estimate MES, which consists of the difference between the total cross-section area and the areas of the other tissues. Therefore, as the areas occupied by the other tissues in the leaf blade increase, the area occupied by the mesophyll is reduced.

Leaves harvested in the pastures under lower grazing intensities (rLAI = 1.8 and 2.3) showed a higher incubation residue after 12, 48, 72, and 96 h of incubation when compared with those of treatments rLAI 0.8 and 1.3 at the same incubation times (Figure 2). Leaves originating from pastures managed under a lower grazing intensity were likely older, due to the less intense grazing, and thus showed a higher percentage of sclerenchyma in their cross-section, mainly in pastures managed under a rLAI of 2.3 (Table 1). Differences in leaf tissue degradation are related to the high cell wall content associated with aspects of the anatomical nature of C4 grass species due to their greater proportion of vascular tissue. Studies have shown that the sclerenchyma tissue provides resistance to leaf breakage (Li et al. 2020), and this fact may interfere with the harvest by the animal, due to its preference; moreover, the amount and location of the leaves can also interfere with the digestion of particles.

Another component that interferes directly in the digestion of the leaf components is lignin (Grev et al. 2019), which delays the hydrolysis of epidermis and sclerenchyma cells, preventing the digestion of vascular tissues. Therefore, the lignin present in organs and tissues as a structural component may produce a negative effect on the fiber digestibility, which might have occurred in the samples of pastures managed under higher rLAI. The proportion of mesophyll, in turn, is positively correlated with digestibility, since the mesophyll cells do not have a secondary cell wall, which allows them to be the first to be digested by the rumen microorganisms.

5. Conclusions

Grazing intensities and period of the year alter leaf anatomy, with the lower grazing intensity (rLAI=2.3) and the dry period increasing the proportion of sclerenchyma of Xaraés palisadegrass. The rLAI between 1.3 and 1.8 provide morphogenetic and structural characteristics more consistent with pastures better managed.

There are correlations between leaf anatomy and morphogenic and structural characteristics. Pre-grazing LAI and leaf elongation rate increases are positively associated with abaxial epidermis and vascular tissues.

Grazing intensities alter leaf blade degradability in the rumen. Lower grazing intensity (rLAI 2.3) provides more residue after leaf incubation.

We can recommend that pasture management of Xaraés palisadegrass be done between rLAI 1.3 and 1.8.

Authors' Contributions: BASSO, K.C.: conception and design, acquisition of data, and analysis and interpretation of data; GALZERANO, L.: acquisition of data and critical review of important intellectual content; DA SILVA, W.L.: analysis and interpretation of data, drafting the article, and critical review of important intellectual content; RUGGIERI, A.C.: critical review of important intellectual content; REIS, R.A.: conception and design, analysis and interpretation of data, and critical review of important intellectual content. All authors have read and approved the final version of the manuscript.

Conflicts of Interest: The authors declare no conflicts of interest.

Ethics Approval: The study was approved by the Animal Research Ethics Committee of the UNESP with number 021119/11.

Acknowledgments: The authors Kelen Cristina Basso and Leandro Galzerano would like to thank Fundação de Apoio à Pesquisa do Estado de São Paulo (FAPESP, process number 09/18482-6) and Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for the scholarships. The authors would like to thank the anonymous reviewers for their valuable comments and suggestions to improve the quality of this paper.

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Received: 10 May 2021| **Accepted:** 22 January 2022 | **Published:** 31 March 2023