http://journal.unoeste.br/index.php/ca/index
DOI: 10.5747/ca.2019.v15.n3.a295
ISSN on-line 1809-8215

Colloquium Agrariae

Submetido: 03/08/2018 Revisado: 02/05/2019 Aceito: 05/05/2019

Bermuda grass foliar chlorophyll content influenced by luminosity and substrate

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Abstract
Bermuda grass (*Cynodon* spp.) is the best suited for ornamental and sportive lawns, widely used in fields of soccer, polo, golf, tennis, baseball and lining among ornamental components of a garden. Regardless of use, a very important characteristic for a good quality lawn is the intensity of green coloration that it presents. The objective of this work was to evaluate the influence of luminosity and different substrates on Bermuda grass (*Cynodon dactylon* x *C. transvaalensis*) chlorophyll a and b contents. The experimental design was completely randomized in a 5 x 4 factorial scheme (substrates x luminosities), in completely randomized design with 20 treatments and 3 replicates. The substrates were: S1 = Soil, S2 = Soil + sand (2:1), S3 = Soil + organic matter (1:1), S4 = Soil + organic matter + sand (2:1:1) and S5 = organic matter + sand (3:1), in four luminosity conditions: full sun, 30%, 50% and 80% of shading for six months. We evaluated chlorophyll *a* and *b*. After 30 DAP the levels of chlorophyll content decreased in relation to shading increased the opposite was found at 180 days after grass planting, higher levels of chlorophyll content were observed as shading increased. The substrate formulations used have not interfered at leaves chlorophyll *a* and *b* content
Keywords: Chlorophyll *a* and *b*; *Cynodon dactylon* x *C. transvaalensi;* shading levels.

Conteúdo de clorofila foliar influenciado pela luminosidade e substrato em grama Bermuda

Resumo
A grama Bermuda (*Cynodon* spp.) é uma das espécies de forração mais utilizadas em campos de futebol, polo, tenis, beisebol, além de fazer parte de components ornamentais em jardins. Além de qualquer uso, uma característica muito importante de qualquer gramado é a colocação verde que possui. Objetivou-se com o experimento avaliar a influência da luminosidade e de diferentes substratos no teor de clorofila das folhas e avaliação visual do sistema radicular de grama bermuda (*Cynodon dactylon* x *C. transvaalensis*). O delineamento experimental foi inteiramente casualizado em esquema fatorial 5 x 4 (substratos x luminosidades), em delineamento inteiramente casualizado com 20 tratamentos e 3 repetições. Os substratos foram: S1 = Solo, S2 = Solo + areia (2:1), S3 = Solo + matéria orgânica (1:1), S4 = Solo + matéria orgânica + areia (2:1:1) e S5 = Matéria orgânica + areia (3:1), em quatro condições de luminosidade: pleno sol, 30%, 50% e 80% de sombreamento, durante seis meses. Foram avaliados: clorofila a e b. Após 30 dias os níveis de conteúdo de clorofila diminuíram conforme aumentou o sombreamento e o oposto foi encontrado aos 180 dias do plantio da grama, em que maiores teores de clorofila ocorreu com aumento do sombreamento.
Palavras-chave: Clorofila *a* e *b ; Cynodon dactylon* x *C. transvaalensis*; níveis de sombreamento.

Introduction

Bermuda grass (*Cynodon* spp.) is the best suited for ornamental and sportive lawns, widely used in fields of soccer, polo, golf, tennis, baseball and lining among ornamental components of a garden. It costs on average 50% more than emerald grass, due to its better quality, lower supply in the market, forming dense lawns with thin leaves (JIMÉNEZ, 2008; CANAL RURAL, 2013).

Following recommendation of Brazilian Organizing Committee Cup 2014 (2009), Bermuda grass hybrids such as Celebration and Tifway 419 were implanted at soccer arenas because they had greater resistance to trampling and faster regeneration, as well as greater facilitating ball bearing and cushioning players impact. Thus, cultivar Tifton/Tifway 419/ITG 6 was used at arenas of Ceará, Bahia, São Paulo, Paraná, Santa Catarina, Rio Grande do Sul and São Januário in Rio de Janeiro. On the other hand, Celebration was used at Goiás, Minas Gerais and Engenhão in Rio de Janeiro.

According to Jimenez (2008), the main advantages of Bermuda grass are: excellent tolerance to water scarcity, high efficiency in water use, salinity tolerance, intense growth, fast establishment, traffic tolerance and tolerance to low cuts, as presented by Jiménez (2008). However, it also has some disadvantages, such as non-tolerance to low-light conditions (shading), high demands on nitrogen fertilizers, low tolerance at low temperatures and compacted soils with poor drainage.

Regardless of use, a very important characteristic for a good quality lawn is the intensity of green coloration that it presents. One of factors linked to plants photosynthetic efficiency, and consequently, to growth and adaptability to various environments is the content of chlorophyll and carotenoids. Regardless the total concentration of these pigments, the ratio between them and between chlorophylls *a* and b changes as a function of light intensity. In general, chlorophyll and carotenoids tend to increase with light intensity reduction (SCALON *et al.*, 2003).

Santos *et al.* (2016), evaluating different substrates at ornamental lawn development showed that treatments with soil and organic matter (2:1); soil, sand and organic matter (2:1:1); and sand and organic matter (3:1) presented better results, with higher production in fresh and dry matter, as well as higher chlorophyll content, showing the benefit of adding organic matter to substrate for grasses cultivations.

In a study with emerald grass, Santos and Castilho (2016), concluded that with organic matter as a substrate component, there were better results regarding the nitrogen and chlorophyll content of leaves.

The objective of this work was to evaluate the influence of luminosity and different substrates on leaf chlorophyll content and visual evaluation of Bermuda grass (*Cynodon dactylon* x *C. transvaalensis*) root system.

Materials and Methods

The experiment was conducted in a region whose clime was classified by Köppen as Aw type, characterized by tropical humid with rainy season in summer and dry in winter. The average annual precipitation is around 1300 mm, distributed from October to March. The relative air humidity varies from 70% to 80% (annual average) and annual temperature mean is approximately 23.5 °C (BINOTTI *et al.*, 2014).

The cultivar Tifway 419 (interspecific hybrid of C. dactylon x C. transvaalensis) was implanted in black plastic containers (47.5 x 17.5 cm – top; 41.5 x 11.3 cm – bottom; 15, 5 cm high, 8.46 L volume), using donated carpets from Itograss®, located in municipality of Pereira Barreto-SP.

The experimental design was factorial scheme 5 x 4 (substrates x shade levels) with three replicates. Plots were submitted to five substrate combination: S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) and S5 = organic matter + sand (3:1). The luminosity levels tested were: full sun, 30%, 50% and 80% shading. The substrates were defined according to results obtained by Santos e Castilho (2016). Structures were made of 1/4" iron rebar with 1 x 1 x 1 m (height x width x length) of dimension where black shade polypropylene shading screens were fixed.

The soil used for substrates composition is classified as Dystrophic Red Latosol, sandy loam-clay texture according to the Brazilian Soil Classification System (EMBRAPA, 2013). The organic matter was obtained through a composting process of vegetal residues with cattle manure. Thick sand with 0.6 to 2.0 mm particle size was purchased from building materials store. After substrates preparation chemical analysis were performed according methodology described by Raij *et al.* (1987) and Embrapa (2017) and the results is shown at Table 1.

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Table 1. Fertility analysis of used substrates in the experiment.

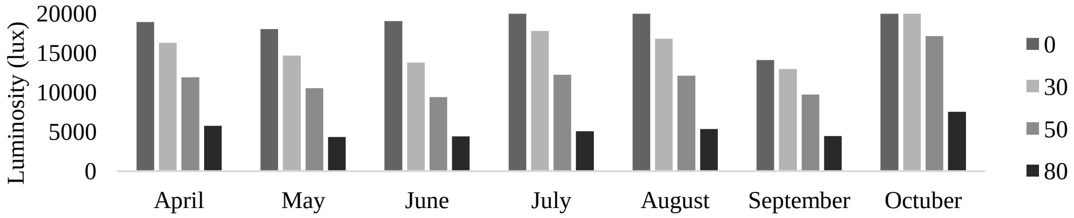
Subs.	P - resin	MO	pH	K	Ca	Mg	H+Al	Al	SB
	mg dm^{-3}	g dm^{-3}	$CaCl_2$			mmol$_c$ dm^{-3}			
S1	9.0	20.0	4.4	1.8	15.0	9.0	47.0	4.0	25.8
S2	10.0	17.0	4.6	1.4	11.0	7.0	38.0	3.0	19.4
S3	892.0	36.0	5.8	14.4	177.0	64.0	26.0	0.0	255.4
S4	476.0	25.0	5.4	7.3	89.0	37.0	28.0	0.0	133.3
S5	976.0	36.0	6.3	9.7	180.0	65.0	16.0	0.0	254.7
I.V.	>120.0		6.0 - 7.0	> 6.0	>7.0	>8.0			
Subs.	**S-SO$_4$**	**CEC**	**V**	**m**	**B**	**Cu**	**Fe**	**Mn**	**Zn**
	mg dm^{-3}	mmol$_c$ dm^{-3}	%				mg dm^{-3}		
S1	8.0	72.8	35.0	13.0	0.23	1.1	32.0	16.8	0.5
S2	5.0	57.4	34.0	13.0	0.21	0.9	34.0	21.5	0.6
S3	61.0	281.4	91.0	0.0	1.26	3.3	111.0	18.6	10.0
S4	35.0	161.3	83.0	0.0	0.75	2.0	80.0	16.6	5.1
S5	58.0	270.7	94.0	0.0	1.02	2.0	56.0	13.0	9.5
I.V.	>10.0		>70.0	< 5.0	> 0.60	>0.8	>12.0	> 5.0	> 1.2

I.V. = ideal values; S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) e S5 = organic matter + sand (3:1).

Irrigation management was daily performed, manually, receiving water until saturation to ensure field capacity of each substrate. At 57 days after planting (DAP) a reduction in grass development was verified, therefore, commercial fertilizer Forth Jardim® (N – 13%; P – 5%; K – 13%; Ca – 1%; Mg – 1%; S – 14%; B – 0.06%; Cu – 0.05%; Fe – 0.20%; Mn – 0.10%; Mo – 0.005%; Zn – 0.20%) was applied, following manufacturer recommendations (100 g m^{-2} and irrigating afterwards). Irrigation and fertilization were performed for did not interfere to results.

The luminosity was determined daily with luximeter placed on the lawns within the structure as shown at Figure 1.

Figure 1. Average luminosity (lux) at each level of shading during experimental period.



At 30 (May) and 180 (October) DAP leaves chlorophyll content was evaluated, based on methodology described by Linder (1974) and Whitham *et al.* (1971).

The means were submitted to mean test and when significant Tukey test was applied at 5% of probability level and regression analysis performed by SISVAR program (FERREIRA, 2011).

Results and Discussion

The average levels of chlorophyll *a* content are shown in Table 2. Neither at 30% or 50% of shading presented means statistically different. It may be seen only in full sun where substrate S3 (soil + organic matter (1:1)) presented the highest mean and differed statically from all others substrates. At 80% of shading substrate S4 (soil + organic matter + sand (2:1:1)) differed statistically from S1 (soil).

Table 2. Chlorophyll *a* content mean(mg gmf^{-1}) in leaves of Bermuda grass submitted to shade treatments and substrates at 30 and 180 days after planting (DAP).

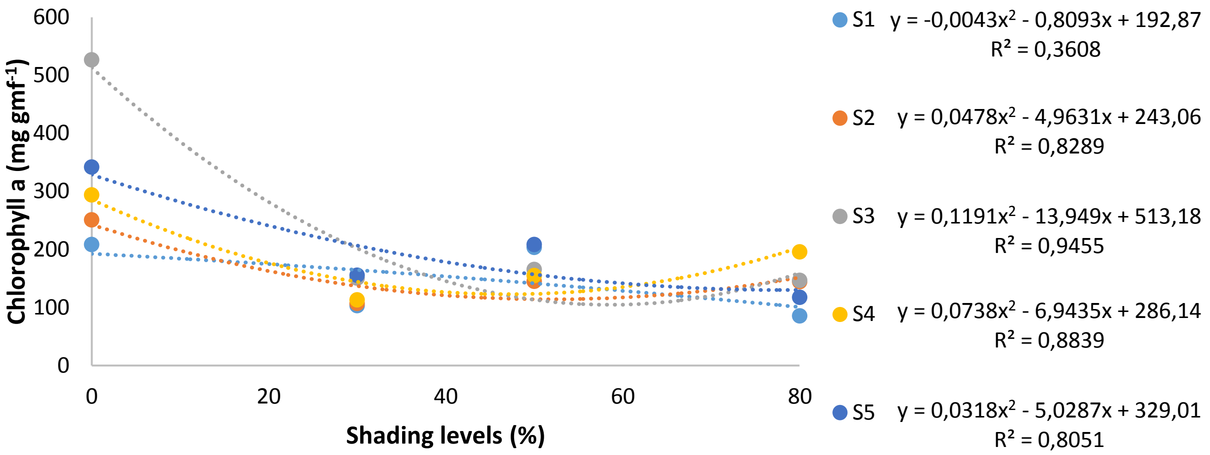
Substrates	30 DAP			
	Shading levels			
	0	30	50	80
S1	208.26 C	103.21 A	203.28 A	85.42 B
S2	250.67 BC	103.76 A	144.91 A	144.48 AB
S3	526.09 A	150.28 A	165.14 A	146.70 AB
S4	293.98 BC	112.93 A	154.96 A	195.46 A
S5	341.89 B	155.24 A	208.53 A	117.22 AB
CV			23,23	

Means followed by same capital letter in the column do not differ statistically from each other at the 5% probability level by Tukey test. [S1 = Soil; S2 = Soil + sand (2: 1); S3 = Soil + organic matter (1: 1); S4 = Soil + organic matter + sand (2: 1: 1); and S5 = Organic matter + sand (3: 1)]

Figure 2 shows regression curves of chlorophyll *a* content at first evaluation (30 DAT), and we may observe quadratic behaviour for all treatments. Deriving equations it is possible to observe the lowest values between 47.04% and 94.10% respectively to substrate S4 and S1, with average value of 58.56% of shading which means chlorophyll *a* reduction when shading levels increased.

At 30 DAP it was possible to observe substrate S1 (soil) obtained the highest value when submitted to full sun and at 50% of shading, while for other substrates the highest value was observed in full sun. In addition to this, in all substrates the lowest value of chlorophyll *a* occurred at the highest level of shading (80%) (Figure 2). Comparing the extreme light treatments, full sun and 80%, both differ statistically in all substrates being higher in full sun (Figure 2). The highest Mg contents (Table 1) provided by organic matter addition in substrates S3 (soil + organic matter (1:1)), S4 (soil + organic matter + sand (2:1:1)) and S5 (S5 = organic matter + sand (3:1)) may have favoured chlorophyll *a* content increase, being this element one of the constituents of this pigment (STREIT *et al.*, 2005).

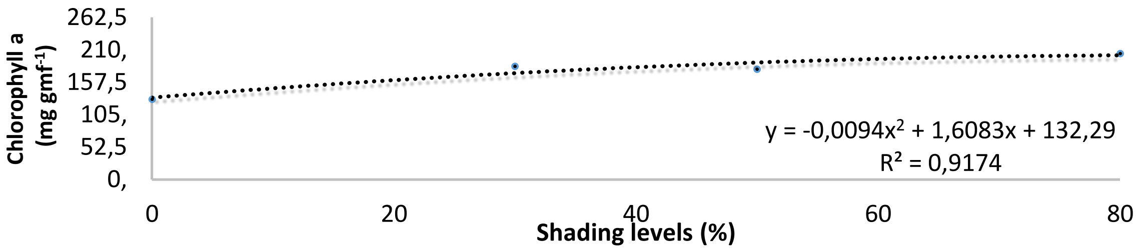
Figure 2. Chlorophyll *a* content (mg gmf^{-1}) in Bermuda grass leaves according to different shading levels 30 days after planting (DAP). S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) e S5 = organic matter + sand (3:1).



At the last evaluation (180 DAP) quadratic behaviour is shown at chlorophyll *a* regression curve (Figure 3). It is observed tendency to increase chlorophyll *a* content as it increases shading level by addition of organic matter rich of Mg, as explain previously at 30 DAP. Deriving equation was obtained the maximum value of shading, which is 85.55%.

At final evaluation (180 DAP), S1, in 30% and 80% shading levels presented the highest chlorophyll *a* content, and the lowest value was observed at full sun. Substrates S2 (soil + sand (2:1)) and S3 the highest content was observed in 80% of shading, and the lowest value in the full sun (Figure 3)).

Figure 3. Chlorophyll *a* content (mg gmf^{-1}) in Bermuda grass leaves according to different shading levels 180 days after planting (DAP). S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) e S5 = organic matter + sand (3:1).



At 180 DAP, chlorophyll *a* tended to be different from observed at 30 DAP. In the first evaluation, there was reduction on content as shading levels increased in the other hand at last evaluation there was content increase for the same condition (Figures 1 and 2).

Baruch and Guenni (2007) verified that there was increase in chlorophyll *a* concentration at moderate levels of shading (40%) in *Brachiaria brizantha* and *B. decumbens*. Páez, González and Pereira (1994) verified significant difference between shading presence and absence, with higher concentration of chlorophyll *a* when *Panicum maximum* was submitted to shading, results different from observed in the present study.

The highest concentration of chlorophyll *a* in treatments with higher shading levels was also observed by Oliveira *et al.* (2013) in two species of grasses (*Andropogon gayanus* cv. 'Planaltina' and *Panicum maximum* cv. 'Tanzania'), justifying this fact as a plant response to a better use of light in shaded environments.

The average chlorophyll *b* content in each treatments is shown in Table 3. At first evaluation of chlorophyll *b* content (Table 3), substrates S3 and S5 presented higher content in full sun. At 30% shading level, substrate S1 despite having a lower content of Mg (Table 1) in its composition, showed the highest content of chlorophyll *b*, but did not differ from substrates S3 and S5 (higher Mg content – Table 1). As with chlorophyll a, the chlorophyll *b* content may also have been influenced by nutrients increase due to fertilization.

Table 3. Chlorophyll *b* content mean(mg gmf^{-1}) in leaves of Bermuda grass submitted to shade treatments and substrates at 30 and 180 days after planting (DAP).

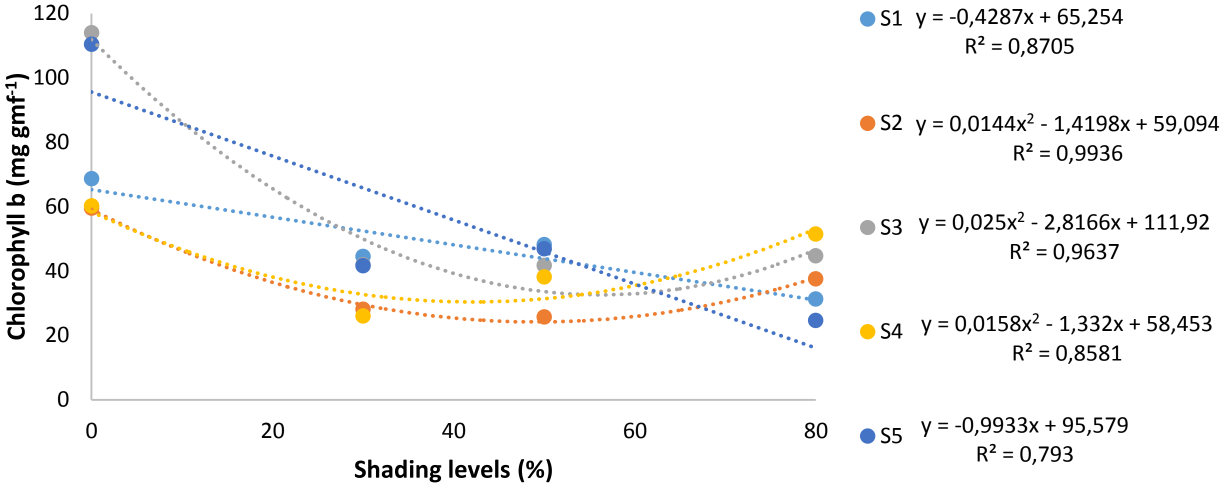
Substrates	30 DAP			
	Shading levels			
	0	30	50	80
S1	59.75 B	66.19 A	53.43 A	31.21 A
S2	59.46 B	30.77 B	25.66 A	35.55 A
S3	109.00 A	41.87 AB	41.68 A	44.63 A
S4	60.13 B	28.99 B	38.11 A	51.44 A
S5	93.33 AB	41.59 AB	46.88 A	24.60 A
CV	30.16			

Means followed by same capital letter in the column do not differ statistically from each other at the 5% probability level by Tukey test. [S1 = Soil; S2 = Soil + sand (2: 1); S3 = Soil + organic matter (1: 1); S4 = Soil + organic matter + sand (2: 1: 1); and S5 = Organic matter + sand (3: 1)]

Figure 4 shows regression curves of each treatment, all curves have quadratic behaviour, and only substrate S1 has a positive curve. Deriving its equation it was possible to find the maximum shading point equal to 20.36% and minimum point varied between 42.65% and 80.54% for substrates S4 and S5, respectively, with an average value of 50.18%.

At 30 DAP substrate S1 had the highest content when submitted to 30% of shading and the lowest value to 80% (Figure 4). The highest value was observed when submitted to the full sun, for substrates S2, S3 and S5. In substrate S2 the lowest content occurred at 50% of shading, in substrates S3 and S5 the presence of shading was responsible for the lowest value. Substrate S4 showed no difference between levels of shading. In relation to shading levels, only full sun and 30% of shading had difference between substrates, and in full sun substrate S3 presented the highest value and substrates S1, S2 and S4 presented the lowest, differing from each other. However, at 30% shading level, substrate S1 presented the highest value of chlorophyll *b* content, and the substrates S2 and S4 had the lowest values for this trait (Figure 4).

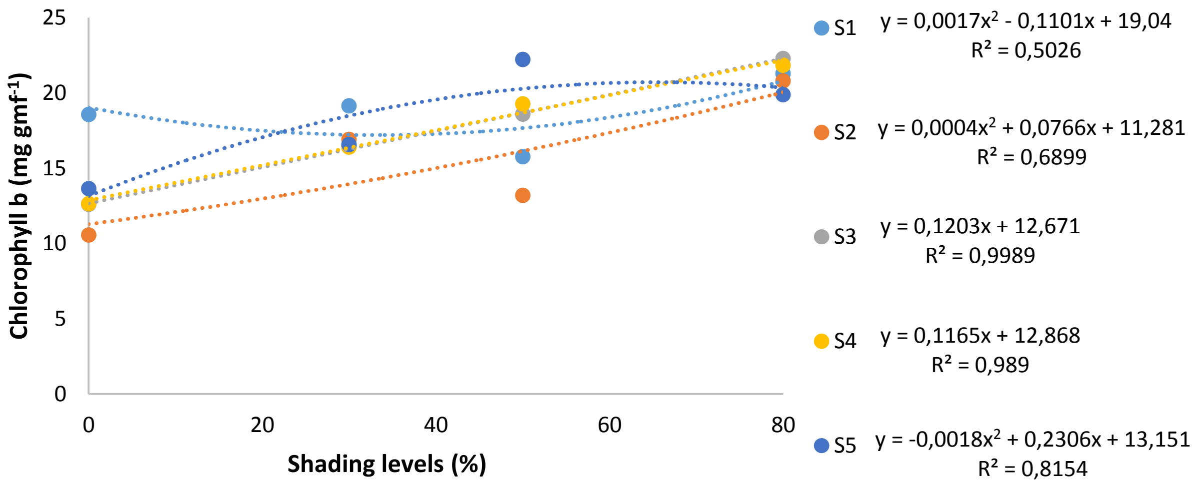
Figure 4. Chlorophyll *b* content (mg gmf^{-1}) in Bermuda grass leaves according to different shading levels 30 days after planting (DAP). S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) e S5 = organic matter + sand (3:1).



Comparing chlorophyll type behaviour, chlorophyll *b* content, as observed for chlorophyll *a*, decreases as shading levels increase (Figures 2 and 4).

The regression curves (Figure 5) show linear behaviour for substrates S3 and S4 and for others substrates quadratic behaviour and for all substrates there was a trend towards higher levels of chlorophyll *b* for more intense levels of shading. Deriving equations, it was possible to find the minimum and maximum points of some curves, the minimum point of shading presented by substrate S1 was 32.38%, and the maximum shading point in substrates S2 and S5 was, respectively, 95.75 % and 64.06% (Figure 5).

Figure 5. Chlorophyll *b* content (mg gmf^{-1}) in Bermuda grass leaves according to different shading levels 180 days after planting (DAP). S1 = soil; S2 = soil + sand (2:1); S3 = soil + organic matter (1:1); S4 = soil + organic matter + sand (2:1:1) e S5 = organic matter + sand (3:1).



The chlorophyll *a* content at the end of experiment had similar behaviour to chlorophyll *b* content, in other words, as shading level increased, chlorophyll *a* and *b* content also increased (Figure 2 and 4).

The result differs from found by Pinto *et al.* (2007), evaluating *Aloysia gratissima* (holy grass), which verified significant difference between shade levels, being the highest concentrations of chlorophyll *b* observed at more intense shading levels. Plants belonging to eudicots group, present structural differences and metabolic cycles (C3) consequently their adaptation to shading, being more apt to this environmental condition (SANTOS *et al.*, 2015), as Bermuda grass belongs to group of C4 plants, behaving differently than eudicots.

The behaviour of chlorophyll *b* content reduction with shading increase was observed in eudicotiledonous species *Cariniana legalis* (jequitibá-rosa), by Rego and Possamai (2011). Similar was found in eudicots, as observed by Engel and Poggiani (1991) in seedlings of native species (*Amburana cearensis*, *Zeyhera tuberculosa*, *Tabebuia avellaneda* and *Erythrina speciosa*).

The increase in chlorophyll *b* levels at the most intense levels of shading occurs because chlorophyll *b* is an accessory pigment, acting in order to increase absorption spectrum that chlorophyll *a* may capture (CASTRO *et al.*, 2009).

Conclusions

After 30 DAP the levels of chlorophyll content decreased in relation to shading increased the opposite was found at 180 days after grass planting, higher levels of chlorophyll content were observed as shading increased.

The substrate formulations used have not interfered at leaves chlorophyll *a* and *b* content

Acknowledgments

Authors acknowledge Coordination of Superior Level Staff Improvement (CAPES) for financial support and Itograss® for grass donation.

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