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Growth and bromatological characteristics of *Brachiaria decumbens* and *Brachiaria ruziziensis* under shading and nitrogen[1]

Características bromatológicas e de crescimento de *Brachiaria decumbens* e *Brachiaria ruziziensis* sob sombreamento e nitrogênio

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ABSTRACT - This study aimed to evaluate the productive and qualitative characteristics of *Brachiaria decumbens* and *Brachiaria ruziziensis* subjected to three levels of artificial shading (0, 36 and 54%) and four nitrogen (N) doses (0; 50; 100 and 150 mg dm^{-3} soil), using completely randomized design with a factorial scheme 2 x 3 x 4, with three replications. The dry matter production (DMP), the number of tillers per pot, root weight and crude protein (CP) and neutral detergent fiber (NDF) contents were evaluated. Both grasses responded quadractly to N, but *B. ruziziensis* presented greater production under the two highest N doses. The tillers density increased with N dose and was reduced under shading. The root weight increased with N dose, linearly in the full sun and quadractly in the shade. The shading and N showed a positive influence on CP contents. For NDF content, was observed reduction with increase of N dose. The NDF content of *B. decumbens* increased with shading levels; for *B. ruziziensis*, the greatest value was observed under intermediate shading level. The N fertilization is an important strategy to improve DMP, tillers density and CP content, for both grasses. However, intense shading should be avoided, as it reduces tillering and root weight, which may threat pasture persistence.

Key words: Crude protein. Fibrous fraction. Silvopastoral systems. Tillering. Tropical grasses. *Urochloa*.

RESUMO - Objetivou-se avaliar características produtivas e qualitativas de *Brachiaria decumbens* e *Brachiaria ruziziensis* submetidas a três níveis de sombreamento artificial (0, 36 e 54%) e quatro doses de nitrogênio (N) (0, 50, 100 e 150 mg dm^{-3} de solo), utilizando-se o delineamento inteiramente casualizado, em esquema fatorial 2 x 3 x 4, com três repetições. Avaliou-se a produção de matéria seca (PMS), o número de perfilhos, a massa de raiz e os teores de proteína bruta (PB) e de fibra em detergente neutro (FDN). Ambas as gramíneas responderam quadraticamente ao N, mas a *B. ruziziensis* foi mais produtiva nas duas maiores doses de N. A densidade de perfilhos aumentou com a dose de N e diminuiu com o sombreamento. O peso de raiz aumentou com a dose de N, linearmente no sol pleno e quadraticamente na sombra. O sombreamento e o N influenciaram positivamente os teores de PB. Para os teores de FDN, houve redução com o aumento da dose de N. Para a *B. decumbens*, os teores de FDN aumentaram com o sombreamento; para *B. ruziziensis* o maior valor foi observado com sombreamento intermediário. A fertilização nitrogenada é uma importante estratégia para o aumento da PMS, do número de perfilhos e dos teores de PB da forragem, para ambas as gramíneas. Contudo, o sombreamento severo deve ser evitado, pois reduz o perfilhamento e o peso de raiz, o que pode comprometer a persistência da pastagem.

Palavras-chave: Fração fibrosa. Gramíneas tropicais. Perfilhamento. Proteína bruta. Sistemas silvipastoris. *Urochloa*.

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INTRODUCTION

The increased interest in the use of silvopastoral systems (SPS) has resulted in an increase in the demand for studies about the behavior of forage plants in shaded environments. According to Castro *et al*. (1999), the distribution of dry matter between different organs of plants is a process strongly influenced by environmental conditions, in addition to genetic differences among species. Therefore, the choice of forage species to be used is important for the success of the system.

There is an association between carbon and nitrogen in the plant, and nitrogen plays an important role in several morphogenic characteristics involving leaf and tiller dynamics. Therefore, studies are necessary to evaluate the potential response of tropical grasses to nitrogenous fertilization associated with the shading effect (GARCEZ NETO *et al*., 2002).

Although tropical forages do not present nutritional quality comparable to that of temperate grasses, their high dry matter (DM) production potential may result in high animal productivity, and nitrogen fertilization may have a positive effect on the quality of the forage produced (CORRÊA *et al*., 2007).

Research carried out in SPS in Atlantic Forest areas in the Southeast region of Brazil has clearly demonstrated the potential of these systems to improve soil fertility in pastures, which results in an increase in the growth of grass, as well as in its nutritive value, in addition to promoting thermal comfort for grazing animals. However, factors such as the level of shade, environmental conditions, tolerance of forage species to shading and characteristics of the trees can influence pasture responses to shading (CARVALHO *et al*., 2007).

In shaded pastures, a very important aspect is the forage quality, which directly influences animal production. Among the effects of shading on grasses, the most consistent is an increase in the crude protein content. Wilson (1996) discussed the causes of the positive effects of shade on growth and nitrogen accumulation in grasses and concluded that these effects are due to an acceleration in the organic matter release and nitrogen cycling. The environmental factors modified by shading have a profound effect on forage quality, since dry matter digestibility and nutrient content are determined by the morphological, anatomical and chemical composition of the forage plant (LIN *et al*., 2001).

The aim of this study was to evaluate the main morphological changes, dry matter production and chemical composition in two forage grasses under nitrogen fertilization and shading, aiming toward their use in SPS.

MATERIAL AND METHODS

The work was carried out in the experimental area at the Dairy Cattle Research Center (Embrapa) in Juiz de Fora, MG. The geographical coordinates are 21°41’20” S latitude, 43°20’40” W longitude, and the average altitude is 678 m. The climate of the region according to Köppen’s classification fits the Cwa type (mesothermal).

Two grasses (*Brachiaria decumbens* cv. Basilisk and *Brachiaria ruziziensis* cv. Kennedy) were studied under three shading levels (0, 36 and 54%) and four nitrogen doses (0, 50, 100 and 150 mg dm^{-3} of soil), which were divided into two equal applications. A completely randomized experimental design was used, with a three-way factorial scheme (2 × 3 × 4) and three replicates.

The shading was implemented using polypropylene screens with different degrees of radiation transmission, which were installed 2 m above the benches. The screens were also installed on the side to avoid sun light incidence in the morning and afternoon. The degrees of shading were calculated based on the photosynthetically active radiation measured under full sun conditions and under shade using a Decagon ceptometer (Accupar model LP 80). The readings of incident radiation were made between 11 and 14 hours for each shading level during the experimental period.

Seeds of *B. decumbens* and *B. ruziziensis* were germinated in plastic boxes on commercial substrates composed of the ground bark of *Pinus* spp. After 20 days of growth, the seedlings were transplanted to tubes of 35 cm^3 containing the same substrate. Approximately 40 days later, three seedlings of each species were transplanted into plastic pots containing 5.0 kg of a mixture composed of soil (Red-Yellow Latosol) and sand at a 4:1 ratio. After the addition of limestone at a dose equivalent to 2 t ha^{-1}, the soil had the following chemical composition: 4.4 mg dm^{-3} P (Mehlich-1), 47 mg dm^{-3} K, 0.8 cmol$_c$ dm^{-3} Ca, 0.3 cmol$_c$ dm^{-3} Mg, 1.82 cmol$_c$ dm^{-3} H + Al, 40% base saturation and a pH (H_2O) equal to 6.3.

Standardization cutting was performed using pruning shears at 5 cm above the soil. The plants were submitted to the different shade levels on 03/10/2008.

Nitrogenous fertilizer (urea) was diluted in water according to the recommended doses and applied to the soil. The volume of solution per application was 50 mL per pot. Together with the nitrogen application, potassium and phosphorus were also added at a dose of 50 mg dm^{-3} for both K_2O and P_2O_5. The first application occurred on the day that the plants were submitted to shading, and the second was performed the day after the first cutting of the plants. The plants were irrigated daily in order to maintain good soil moisture conditions.

Two cuts at 35 days of regrowth were performed in April and May 2008. The cuts were made with pruning shears at a height of 5 cm above the ground level.

On the day of each cut, the number of tillers was counted in order to obtain the number of tillers per pot. Afterwards, the plants were harvested, and the samples were taken to the sample preparation laboratory to determine their morphological characteristics (leaf blade; stem + leaf sheath). After weighing, each fraction was dried in a forced-air oven (55 °C; 72 hours). After this period, the samples were again weighed to determine the dry matter content. The addition of the leaf blade and stem weights was used to obtain the total dry matter weight. The roots were washed to remove the soil, weighed and dried in a forced-ventilation oven (55 °C; 72 hours) to determine the dry matter content.

Chemical composition analysis was carried out at the Animal Nutrition Laboratory of the Federal Rural University of Rio de Janeiro. The samples of leaf blades were dried (55 ºC; 72 hours), milled in a Wiley mill with a 1 mm sieve and analyzed for the contents of dry matter (DM), crude protein (CP) and neutral detergent fiber (NDF) according to Detmann (2012).

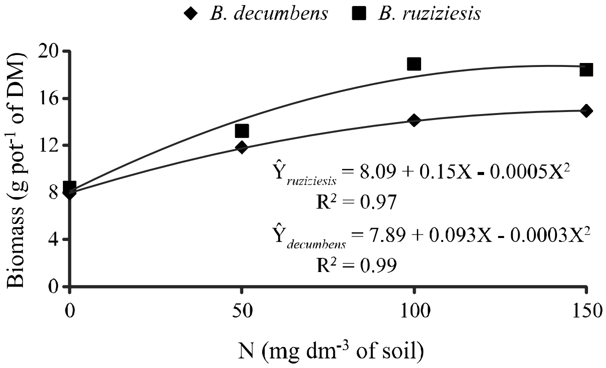
The data were submitted to analysis of variance using the average values of the two harvests. The means were evaluated using the F test (α = 0.05), the SNK test (α = 0.05) and regression analysis for the variables grass species, shade level and N dose, respectively, using the statistical package SAEG (UFV, 2007).

RESULTS AND DISCUSSION

The forage production was influenced by grass (p<0.001), N dose (p<0.001) and the grass × N dose interaction (p = 0.030), but no significant effect of shading was observed (p = 0.155). The values were similar under the two lower N doses but were different when 100 and 150 mg/pot of N was applied. *B. ruziziensis* presented greater forage production than the other grass specie (Figure 1). Bonfim-Silva and Monteiro (2006) observed a maximum value of 17 g pot^{-1} of leaf dry mass for *B. decumbens* when fertilized with a dose of N of 371 mg dm^{-3} of soil. This value was higher than that observed for *B. decumbens* in the present study, which can be attributed to the higher N dose applied by the other authors.

In relation to the N dose, there was a quadratic effect on forage production regardless of the grass species (Figure 1). The positive effect of N fertilization, as well as the quadratic response, were observed in other studies (CORRÊA *et al*., 2007; JOHNSON *et al*., 2001). This result suggests the existence of a point of maximum forage production after which there is no response to N. Nitrogen increases the growth rate of the grass and, consequently, the amount of forage produced per unit of time (SANTOS *et al*., 2009). Johnson *et al*. (2001) reported the existence of a limit after which additional fertilization with N does not promote an increase in forage production.

Figure 1 - Biomass production of *Brachiaria decumbens* and *Brachiaria ruziziensis* in response to nitrogen (N) doses

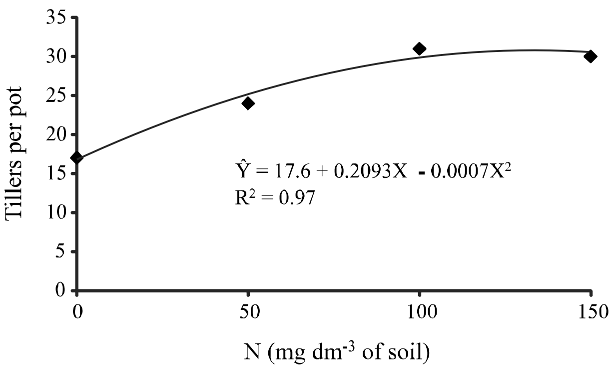


With a progressive increase in external nutrient availability, the absorbed N is invested in the synthesis of metabolically active foliar structures (leaf area), which leads to an increase in forage production. The use of higher N doses (100 and 150 mg dm^{-3}) resulted in a more pronounced increase in the production of *B. ruziziensis*, indicating its greater efficiency in the use of N when compared to *B. decumbens*.

No effect of shading on forage production was observed. DeBruyne *et al*. (2011) also did not observe an effect of up to 50% shading on the production of cold-season forages under natural and artificial shading. Forage production was influenced only when the plants were submitted to 70% shading. The fact that shading did not affect production indicates that these species present a relative tolerance to reduced luminosity, which is possible due to the development of some morphophysiological mechanisms (FERNÁNDEZ; GYENGE; SCHLICHTER, 2004). Among these mechanisms, the change in the biomass allocation pattern between above and belowground structures is important. Martuscello *et al*. (2009) stated that the satisfactory forage production of *B. decumbens* in environments with shade of 50% was due to the partitioning of nutrients to the aerial part of the plant to the detriment of the roots. Other changes in the plant in response to shading can be observed, such as changes in canopy architecture, the angle of inclination of leaves and specific leaf area (FERNÁNDEZ; GYENGE; SCHLICHTER, 2004).

The number of tillers was influenced by the N dose ($p<0.001$) and by the interaction ($p = 0.006$) of shading × grass. A quadratic response was observed for the number of tillers as a function of the N dose (Figure 2). This result reveals the importance of N in the production of new tillers, which can result in increased forage production in addition to contributing to greater soil cover and erosion reduction. Silva *et al*. (2009) observed a similar response when *B. decumbens* and *B. brizantha* cv. Marandu were fertilized with N. The results indicate that the plants are able to respond to nitrogen fertilization up to a point, likely due to i) the limited capacity of the plant to absorb N through its roots, ii) the limitation of other nutrients necessary for the development of the plant, and iii) the limitation of the number of buds determining the tillering potential of the plant. This behavior was also observed by Garcez Neto *et al*. (2002) and Lavres Junior and Monteiro (2003) in *Panicum maximum* cv. Mombaça.

Figure 2 - Tillers number per pot in response to nitrogen (N) doses



According to Oliveira *et al*. (2007), grasses responds positively to nitrogen fertilization, likely due to the stimulus promoted by N in terms of the growth and multiplication of plant cells, since this nutrient constitutes the proteins and cellular nucleic acids. In addition, nitrogen fertilization increases the rate of leaf appearance, which results in a greater number of axillary buds capable of producing new tillers (DE BONA; MONTEIRO, 2010).

When evaluating the grasses at each level of shading, a difference was observed between the grasses only at 36% shading, at which *B. decumbens* presented a larger number of tillers (Table 1). When evaluating the different levels shading within each grass species, differences were observed among shade levels in both grasses. *B. decumbens* showed higher tillering under full sun, with a decrease in the number of tillers only when subjected to the most severe shading. For *B. ruziziensis*, already at 36% shade, there was a reduction in the number of tillers in relation to the full sun condition. These results demonstrate the higher tillering ability of *B. decumbens* compared to *B. ruziziensis* under conditions in which light is partially reduced.

Table 1 - Mean values for tillers number (per pot) in *Brachiaria decumbens* e *Brachiaria ruziziensis,* under shade levels

Grass	Shade (%)		
	0	36	54
B. decumbens	30.3 aA	28.7 aA	24.8 aB
B. ruziziensis	32.6 aA	24.4 bB	24.3 aB
CV = 12.20%			

Means followed by same letter, lowercase in the columns and uppercase in the rows, do not differ according F and SNK tests, respectively ($P>0.05$)

The reduction in the number of tillers with increasing shading shows the importance of light in the emergence of new tillers in pastures, since light activates the basal and axillary buds, promoting the formation of new tillers (MORAIS *et al*., 2006). The reduction in the number of tillers may be due to the decrease in the red: extreme red ratio, which causes a delay in the development of buds without changes in the phyllochron, and the tillering can cease when the red:extreme red ratio reaches values below 0.40 (EVERS *et al*., 2006). Paciullo *et al*. (2007) also observed a reduction in the number of tillers in *B. decumbens* when it was exposed to 54% shading, independent of the season. Gobbi *et al*. (2009) observed a reduction of approximately 50% in the population density of tillers in *B. decumbens* submitted to 70% shading. Martuscello *et al*. (2009) observed a reduction in the number of tillers in *B. decumbens* submitted to shading, with values of 21.7, 17.8 and 8.0 plant^{-1} tillers under full sun and 50 and 70% shading, respectively.

Despite the reduction in the number of tillers caused by shading, no reduction in forage production was observed. Thus, it is possible to consider that the reduction in the number of tillers was compensated for by the size of the tillers. This tiller size/density compensation pattern explains the phenomenon in which forage production remains constant despite variations in the population density and/or average weight of tillers (MATTHEW *et al*., 1995).

The root mass varied with grass ($p<0.0001$), shading ($p<0.001$) and N dose ($p<0.001$). There were also significant interactions grass x shade ($p = 0.019$) and shade × N dose ($p = 0.0048$). The root mass was higher for *B. decumbens* under full sun and 36% shading but did not vary between the grass species at the highest shade level (Table 2).

Table 2 - Average values of root mass (per pot) in *Brachiaria decumbens* and *Brachiaria ruziziensis*, under shade levels

Grass	Shade (%)		
	0	36	54
B. decumbens	13.4 aA	9.1 aB	6.5 aC
B. ruziziensis	9.0 bA	7.5 bB	5.8 aC
CV = 7.68%			

Means followed by the same letters, lowercase in the columns and upper case in the lines, do not differ according F and SNK tests, respectively(P>0.05)

The root mass decreased with an increase in shade level independently of the grass species. Martuscello *et al*. (2009) also observed a reduction in the root weight of *B. decumbens* and *B. brizantha* cvs. Xaraés and Marandu cultivated under full sun or at 50 and 70% shading. According to these authors, the reduction in root mass may be detrimental to the plant, since the roots are important for the uptake of nutrients and water from the soil and for the accumulation of reserves that will facilitate the regrowth of the plant after grazing.

In the evaluation of shade levels within each dose of N, there were differences among all levels of shade at doses of 100 and 150 mg of N dm^{-3} of soil, with plants at full sun presenting the highest root weights and the plants shaded by 54% having the lowest (Figure 3). In the dose of zero N, a difference was observed only between full sun and shaded plants, the latter of which had a lower root mass.

The root mass presented a linear positive response to the different doses of N under full sun. For the shaded plants, there was a quadratic response to the different N doses (Figure 3). These results show that plants submitted to full sun prioritize the stimulation of root growth in response to an increase in N in the soil. The increase in root mass can favor the absorption of water and nutrients by the plant. Pandey *et al*. (2011) attributed\ the higher nitrogen absorption by the species *Panicum maximum* Jacq. and *Brachiaria mutica* (Forssk.) Stapf. to the fact that they present a lower leaf biomass:root biomass ratio, since the presence of nitrogen in the soil would stimulate an increase in the root system, allowing the plant to explore deeper layers of the soil.

For shaded plants, the root mass values in response to N reach a maximum after which there is no increase in root mass. In fact, shaded plants tend to prioritize the growth of the aerial part in relation to the root system in order to compensate for the lower incidence of light.

The CP content was influenced by shading (p<0.001), grass (p<0.001) and nitrogen (p<0.001), and there was also a significant shade × nitrogen interaction (p = 0.010). The highest CP content was obtained for *B. decumbens*, with a mean value of 9.7% against 8.4% for *B. ruziziensis*. Pariz *et al*. (2010) observed mean CP values for *B. ruziziensis* and *B. decumbens* of 10.4 and 6.8%, respectively.

When evaluating the different shade levels at each dose of nitrogen, it was found that there was a difference in CP levels among the shade levels at 100 and 150 mg N dm^{-3} of soil, while for plants with no addition of N and with 50 mg N dm^{-3} of soil, a difference only between plants grown under full sunlight and those under shading was observed (Figure 4).

Figure 3 - Root mass in response to shade levels (%) and N doses

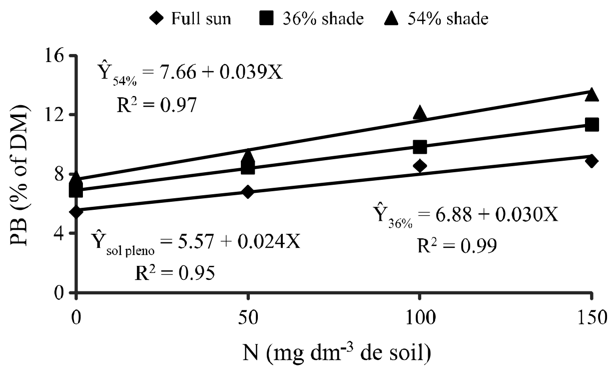
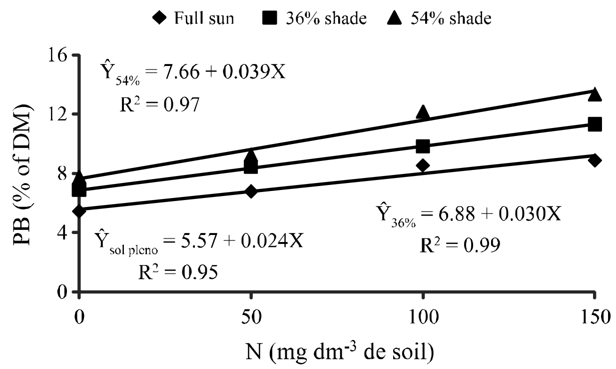


Figure 4 - Crude protein content (CP) in response to shade levels (%) and N doses



Positive linear responses were observed for the CP content at all shade levels but with different N response efficiencies depending on the level of shading (Figure 4). The increases observed were 0.024, 0.030 and 0.039 of a percentage point in CP for each mg of N applied for full sun and shade conditions of 36 and 54%, respectively. This behavior can be explained by the direct effect of nitrogen fertilization on CP levels, as well as by the shading stimulus to increase the N concentration in the plant tissues (MOREIRA *et al*., 2009; PACIULLO *et al*., 2007). It is important to consider that shaded plants undergo morphophysiological changes to suit the environment under reduced luminosity, such as increases in leaf length (PACIULLO *et al*., 2011) and chlorophyll concentration, in order to maximize their photosynthetic potential (LÁZARO, 2007). According to Moreira *et al*. (2009), nitrogen directly participates in the photosynthetic process through its inclusion in the chlorophyll molecule.

Several authors have observed an increase in the CP content of grass leaves grown under shade conditions, artificial or natural (CARVALHO; FREITAS; XAVIER, 2002; CASTRO *et al*., 1999; SOUSA *et al*., 2007; WILSON, 1996), which can be attributed to the higher amount of nitrogen in the leaves of grasses cultivated under low light intensity conditions that accumulates in order to obtain a greater quantity of photosynthetically active compounds for the better use of the incident light.

The NDF content varied as a function of the grass × shade ($p<0.001$), grass × nitrogen ($p = 0.016$) and shade × nitrogen ($p = 0.003$) interactions. By evaluating the levels of shade within each grass species, differences were observed for all levels of shade in *B. decumbens*, whereas for *B. ruziziensis*, there was difference only at the shade level of 36% (Table 3).

In the evaluation of grasses within each shade level, a difference was observed between grasses only when they were submitted to different shading levels. *B. ruziziensis* had a higher NDF content under 36% shad

Table 3 - Average values of neutral detergent fiber (NDF) as a function of grass and shade levels

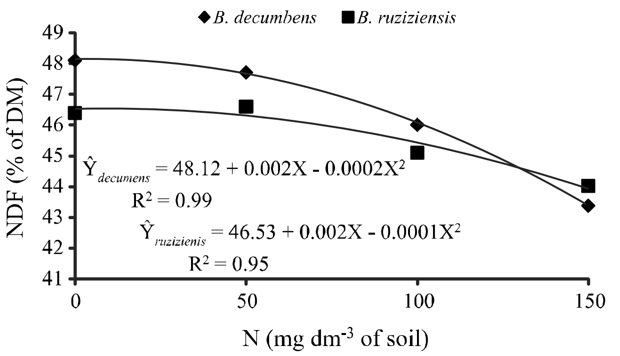
Grass	Shade (%)		
	0	36	54
B.decumbens	44.75 aC	46.16 bB	47.97 aA
B.ruziziensis	44.42 aA	47.10 aB	45.12 bA
CV = 2.34%			

Means followed by the same letters, lowercase in the columns and upper case in the lines, do not differ among the F and SNK tests, respectively (P>0.05)

and a lower NDF content under 54% shade. An increase in the NDF content of plants subjected to shading was also observed by Lin *et al*. (2001) in plants submitted to artificial shading of up to 80% and by Kirchner *et al*. (2010) in temperate grasses under the shade of *Pinus taeda*. However, the results presented in the literature are varied, with reports of decreases in the NDF content of shaded plants (PACIULLO *et al*., 2007) or no change in the NDF content (CARVALHO; FREITAS; XAVIER, 2002; SOUSA *et al*., 2007). This inconsistency in the results can be explained by differences in the ability of plants to adapt to shade as well as differences in forage species, soil fertility, management and time of year.

In the evaluation of the grass species at each dose of N, negative quadratic responses were observed for both grasses. Differences in the NDF content were observed between the grass species only at doses of 0 and 50 mg of N dm^{-3} of soil, with *B. decumbens* presenting the highest values (Figure 5). Increasing the dose of N allowed for faster tissue synthesis and a concomitant increase in plant growth, to the detriment of the deposition of fibrous constituents in the cell wall. This mechanism was more evident for *B. decumbens*, which showed a greater reduction of the NDF content with an increase in the dose of N.

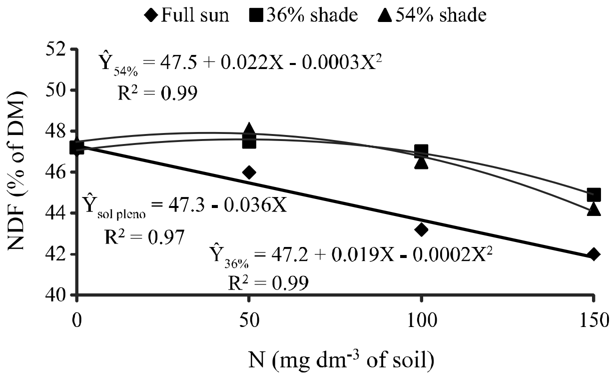
Figure 5 - Neutral detergent fiber (NDF) content of *Brachiaria decumbens* and *Brachiaria ruziziensis* in response to nitrogen (N) doses



By evaluating the shade levels within the N rates, lower NDF contents were observed in plants submitted to full sun when compared to those grown under shading of 36% and 54%. In the absence of N, no differences were observed in the NDF contents among the environments differing in luminosity (Figure 6).

The NDF content of plants grown under full sunlight decreased linearly in response to N dose, while the shaded plants presented a quadratic response, with lower NDF contents observed at the higher N rates. A decrease in NDF content with increased nitrogen fertilization was also observed by Costa *et al*. (2009) in *B. brizantha*. This effect can be explained by the fact that N stimulates the growth of plants and increases the use of available carbohydrates for the formation of cells and protoplasm rather than provoking the thickening of cell walls through the accumulation of these carbohydrates (BLACK, 1968 apud CORRÊA *et al*., 2007). Another hypothesis is related to the occurrence of fibrous fraction dilution, since nitrogen fertilization promoted greater dry matter production in the plants in the present study.

Figure 6 - Neutral detergent fiber content (NDF) in response to shade levels (%) and nitrogen (N) doses



CONCLUSIONS

1. Nitrogen fertilization provides an increase in dry mass, tillering and root mass of *B. decumbens* and *B. ruziziensis*;
2. The grasses showed to be tolerant to the shading, since they did not decrease the dry mass production, when compared to the pasture in full sun. However, more severe shading should be avoided because it reduces tillering and root mass, which could, in the long term, threaten the persistence of pasture;
3. Nitrogen fertilization promotes improvements in nutritive value, considering increases in crude protein and reduction of neutral detergent fiber contents;
4. The positive effect of nitrogen fertilization on the crude protein content is increased by shading.

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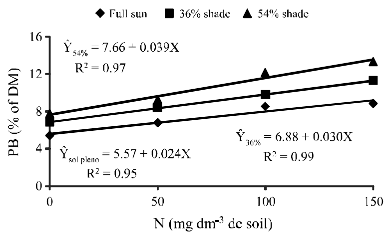
ERRATA

In the article "Growth and bromatological characteristics of *Brachiaria decumbens* and *Brachiaria ruziziensis* under shading and nitrogen" of the authors: Bruna Moscat Faria[2], Mirton José Frota Morenz[3], Domingos Sávio Campos Paciullo[3]*, Fernando César Ferraz Lopes[3] and Carlos Augusto de Miranda Gomide[3], with number of DOI 10.5935/1806-6690.20180060, published in the Revista Ciência Agronômica, volume 49, número 3, July-September, 2018 (Rev. Ciênc. Agron., v. 49, n. 3, p. 529-536, jul-set, 2018), on page 533:

Where to read:

"Figure 3 - Root mass in response to shade levels (%) and N doses"



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"Figure 3 - Root mass in response to shade levels (%) and N doses"