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ORIGINAL PAPER

Effect of increasing densities of *Urochloa brizantha* cv. Marandu on *Eucalyptus urograndis* initial development in silvopastoral system

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Abstract In silvopasture system, the coexistence of eucalyptus seedlings with other species may result in growth reduction, especially during eucalyptus early development. Therefore, studies elucidating how forage species affect the eucalyptus growth can provide important information for their rational management aiming to obtain the maximum gain of the system. The aim of this work was to evaluate the effect of increasing densities of *Urochloa brizantha* cv. Marandu in the early development of *Eucalyptus urograndis*. An experiment was conducted in 20 L pots, in an open and semi-controlled area, during 90 days after planting of eucalyptus. A completely randomized design with four replications was used, in a 6 × 7 factorial system, meaning six evaluation periods and seven densities of *U. brizantha*: 0 (control), 22, 33, 44, 67, 89 and 111 plants m^{-2}. Fortnightly, eucalyptus height, stem diameter and chlorophyll fluorescence (F_v/F_m) were evaluated. At the end of experimental period, the net assimilation rate, stomatal conductance and transpiration rate of eucalyptus plants were determined, in addition to the dry matter of eucalyptus (leaves and stem) and *U. brizantha* (leaves). In coexistence with 111 plants m^{-2}, eucalyptus had reduction of 63.9% on total dry matter and 72.7% on leaf area, compared to the control. From the density of 22 plants m^{-2}, *U. brizantha* negatively interfere significantly the growth of *E. urograndis*. Up to 8 plants m^{-2} there are no reductions greater than 5% in eucalyptus height and stem diameter.

Keywords Eucalyptus · Palisade grass · Competition · Interference

Introduction

Eucalyptus spp. is the most important genus for the forest sector in Brazil, being the main crop supplier of wood and cellulose (Abraf 2013). Due to investments in research of genetic improvement and development of crop management, including an effective management of weeds (Stape et al. 2004; Pereira et al. 2012), eucalyptus culture obtained an annual yield increase of 5.7% between 1970 and 2008, reaching, in 2015, 36 m^3 ha^{-1} a^{-1} in more than 5.6 million hectares of planted area. Thus, Brazil has the eucalyptus plantations with the largest productivity in the world (Iba 2016). Although this crop is predominantly used for pulp and paper production, its consortium with forage plants is often verified in silvopastoral systems (Marques et al. 2015).

The silvopastoral system is characterized by the association of forestry and livestock activities in order to obtain a supplemental production due to the beneficial effect caused by the interaction of their components (Macedo et al. 2001). In this system, the forest species contribute to the comfort and welfare of animals, by providing shade, as well as reducing the impact of the wind and rain on the soil,

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avoiding the deterioration of pasture (Macedo 2009). It also improves the physical properties of the soil, because their roots contributes to increase the water infiltration and water holding capacity, and uses nutrients from deeper soil layers, returning them to the surface through the decomposition of leaves, branches and bark (Menezes et al. 2002).

Urochloa brizantha cv. Marandu, commom named as palisade grass, is widely recommended for grazing plantations in silvopastoral systems, due to shade tolerance (Costa et al. 2000; Ribaski and Rakocevic 2002). In order to maximize the gain in the system, the comprehension of forage plant interference on the eucalyptus initial growth is required, since it will behave as a competitor (a weed plant), resembling to the other species of *Urochloa* that have become weeds in eucalyptus areas in Brazil (Brendolan et al. 2000; Bacha et al. 2016).

The presence of weeds in eucalyptus areas is widely reported in the literature, which due to its competitiveness, have been a challenge in the management of different eucalyptus species (Ellis et al. 1985; Toledo et al. 2000, 2001; Adams et al. 2003; Garau et al. 2008, 2009; Cruz et al. 2010).

In addition to compete with the culture for water, nutrients, light and space, weeds can release allelochemicals into the environment, causing reduction in eucalyptus growth (Pitelli and Marchi 1991; Watt et al. 2003); or they may affect the culture indirectly, hosting pests, diseases, and increasing the risk of fire (Pitelli and Marchi 1991). In this context, several studies have been carried out in order to realize how the different species of weeds affect the different forest species (Sands and Nambiar 1984; Ellis et al. 1985; Caldwell et al. 1995; Adams et al. 2003; Florentine and Fox 2003; Schaller et al. 2003; Coll et al. 2004; Garau et al. 2008; Cruz et al. 2010; Marques et al. 2015).

Eucalyptus plants are sensitive to interference imposed by coexistence with weeds during its initial development period, which comprising from planting until about a year old (Nambiar and Sands 1993; Florentine and Fox 2003; Garau et al. 2009), as this competition can results in physiological changes that adversely affect photosynthetic characteristics of eucalyptus (Santos et al. 2015). Several factors affect the magnitude of the interference, such as: eucalyptus clone resistance to competition (Cruz et al. 2010; Pereira et al. 2013); weed plant species (Torres et al. 2010); weed density (number of weed plants per unit area) and the distance between weeds and eucalyptus plants (Dinardo et al. 2003; Graat et al. 2015; Bacha et al. 2016); and the period of coexistence between the weeds and crop (Toledo et al. 2000). The density is an essential factor to be studied, since even at low intensity (4 plants m^{-2}) some species of weeds lead decreasing of up 55% on the dry matter of *Eucalyptus grandis* stem (Toledo et al. 2001). Therefore, studies in this direction can provide important information to producers and the scientific community in order to guide the management and efficient control of species that become weeds or possible partners in production systems (Marques et al. 2015).

Assuming that high *U. brizantha* densities reduce the growth of *E. urograndis*, this study aimed to evaluate the interference of increasing densities of *U. brizantha* in the early development of *E. urograndis*.

Materials and methods

The experiment was carried out from April to July 2010, in an open and semi-controlled area located at Sao Paulo State University-UNESP/FCAV, Jaboticabal, SP, Brazil (21°14′39.33″S; 48°17′56.41″W). The site has altitude of 592 m, climate Cwa, according to Köppen (1948), subtropical, with rainy summer, dry winter, average annual rainfall of 1552 mm and average temperature of 22 °C (meteorological data in Table 1).

The experiment was conducted during 90 days after planting (DAP) of eucalyptus, and all the plants were planted in pots of 20 L (28 cm in diameter and 32 cm deep) previously filled with substrate formed by soil mixture collected in the surface layer of a red Latosol and sand in the ratio 2:1 (v/v).

Seedlings of a *E. grandis* × *E. urophylla* (*E. urograndis* clone GG100) were used, which had, on average, 90 days old, 20 cm in height, 1.25 mm in diameter and 10–12 leaves.

The experiment was designed in a 6 × 7 factorial scheme, respectively represented by the time of measurements (fortnightly), and weeds density: 0 (control), 1, 2, 3, 4, 5 and 6 plants per pot, corresponding to a density of, respectively, 0, 22, 33, 44, 67, 89 and 111 plants m^{-2}. A completely randomized design with four replications by treatment was used, in which each pot consisted of an experimental unit. All pots were irrigated daily until field capacity and one eucalyptus seedling was transplanted in the center of each pot.

The *U. brizantha* seedlings were obtained by sowing in trays with horticultural substrate in order to standardize the size of the weed plant, being transplanted after having two fully expanded leaves.

Ten days after eucalyptus planting, the *U. brizantha* seedlings were transplanted to the pots at a distance of 5 cm from the eucalyptus, according to the densities proposed for treatments.

As cultural practices, were carried out: a nitrogenous fertilizer at 10 DAP, with 50 mL of 2% urea solution; and a NPK fertilizer (4-14-8; 400 kg ha^{-1}) at 40 DAP. Due to the presence of aphids, an deltamethrin application at a dose of 5.0 g ha^{-1} (EC Decis® 25–200 mL ha^{-1}) was also carried out at 40 DAP.

Fortnightly, until 90 DAP, the eucalyptus plants height and stem diameter were evaluated. On these occasions,

Table 1 Monthly meteorological data of Jaboticabal-SP region, Brazil, 2010

Month	Pressure (hPa)	Tmax (°C)	Tmin (°C)	Tave (°C)	RH (%)	Precipitation (mm)	NRD	Insolation (h)
Apr	944.7	29.2	17.1	22.2	74.6	95.5	07	245.7
May	944.9	27.1	14.1	19.5	72.5	10.6	04	239.9
Jun	948.0	27.4	12.0	18.5	68.3	7.8	02	256.1
Jul	947.8	29.2	13.9	20.4	63.8	0.0	0	278.9

Pressure, atmospheric pressure; Tmax, average maximum temperature; Tmin, average minimum temperature; Tave, average temperature; RH, relative humidity of air; NRD, number of rainy days

were also determined, in the third fully expanded leaf from the plant apex, the fluorescence of chlorophyll $a - F_v/F_m$ (Hansatech, mod. PEA).

At 90 DAP, the end of experimental period, net CO_2 assimilation rate (μmol m^{-2} s^{-1}), stomatal conductance (mol m^{-2} s^{-1}) and transpiration rate (mmol m^{-2} s^{-1}) were evaluated with an infrared gas analyzer (IRGA mod. LI 6400, LiCor®), in the third expanded leaf. The working reference conditions adopted for gas exchange evaluations were: 350 μmol CO_2 mol^{-1}, 9 mmol mol^{-1}, the chamber temperature was set at 25 °C, atmospheric pressure in 1000 kPa, flow rate at 400 μmol s^{-1}, and the photosynthetically active radiation (PAR) at 1000 μmol m^{-2} s^{-1}.

Also at 90 DAP, eucalyptus and weeds were cut close to the soil to leaf area determination (LiCor, mod LI 3100). Leaves and stems were dried in a forced circulation oven (± 70 °C) for 96 h to determine the dry matter mass.

Analysis of variance was performed in 6 × 7 factorial scheme for stem diameter, eucalyptus height and F_v/F_m. We used the multiple regression models for the variables eucalyptus height, stem diameter and chlorophyll fluorescence (F_v/F_m) on original scale, in order to investigate the relationship between the density and time. We use the package rsm from R (R Core Team 2017).

The eucalyptus variables: dry matter, leaf area, net CO_2 assimilation rate, transpiration rate, stomatal conductance, and *U. brizantha* dry matter, were submitted to the analysis of variance by *F*-test. The analysis that showed significance was compared by Tukey test at the level of 5% of probability. All variables suffered Box-Cox transformation to obtain homoscedasticity. The SAS software (Statistical Analysis System, version 9.3) was used for data analysis.

Results

The eucalyptus height, stem diameter and chlorophyll fluorescence (F_v/F_m), had significant interaction between the periods of coexistence and the densities of the forage plant (Table 2).

The eucalyptus height increased over the time, regardless of weed density (Fig. 1). From the 70 DAP on, there was noted an interference of higher weed densities compared to the densities less than 8 plants m^{-2}.

At 90 DAP, the average height of the eucalyptus seedlings that coexisted with 78 plants m^{-2} or more, was 12.3% lower than plants with 8 plants m^{-2} or less. Also, up to 8 plants m^{-2}, it was observed no reduced height growth greater than 5% when compared to the control (Fig. 1).

Table 2 Results of analysis of variance for *Urochloa brizantha* cv. Marandu densities and periods of coexistence on *Eucalyptus urograndis* height (cm), stem diameter (mm) and chlorophyll fluorescence (F_v/F_m)

	Height (cm)	Diameter (mm)	F_v/F_m
Periods of coexistence (days)			
15	45.71 E	3.59 E	0.791 AB
30	50.32 D	4.68 D	0.721 C
45	56.37 C	5.60 C	0.768 B
60	60.62 B	7.07 B	0.700 C
75	65.39 A	8.61 A	0.707 C
90	66.23 A	9.17 A	0.815 A
Densities (plants m^{-2})			
0	60.35 A	7.24 A	0.771 A
22	59.12 AB	7.18 A	0.765 A
33	56.14 C	6.40 BC	0.738 AB
44	53.35 D	5.92 C	0.746 AB
67	58.54 ABC	6.41 B	0.747 AB
89	56.97 BC	5.99 BC	0.729 B
111	57.67 ABC	6.03 BC	0.715 B
F periods	193.28**	551.49**	34.74**
F densities	12.69**	16.23**	4.12**
F periods × densities	2.07**	3.15**	1.69**
CV (%)	5.50	4.51	5.20

Means followed by the same letter do not differ from each other by Tukey test at 5% of probability. **Significant values at 1% of probability by the *F*-test. CV is coefficient of variation. F is *F*-test value

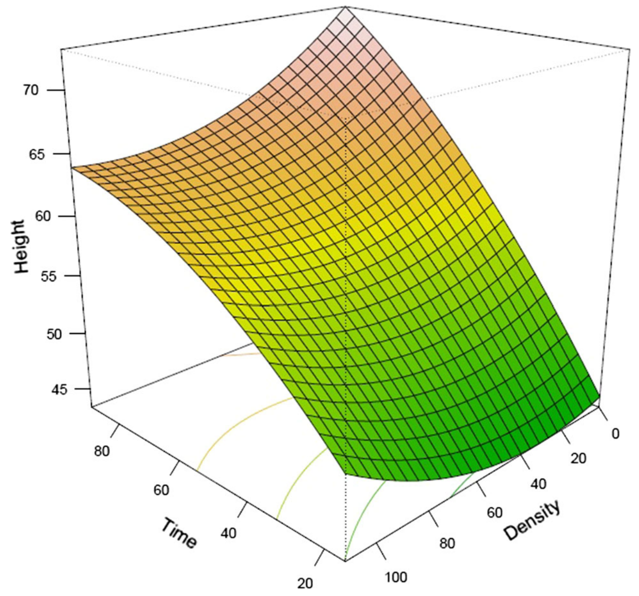

Fig. 1 Effect of increasing densities of *Urochloa brizantha* cv. Marandu on *Eucalyptus urograndis* (clone GG100) height (cm) during 90 days after planting. $Height = 35.9 + 0.5 \cdot Time - 0.03 \cdot Dens - 0.002 \cdot Time \times Dens - 0.002 \cdot Time^2 + 0.001 \cdot Dens^2$

After 90 days, the highest stem diameter values were observed at the lowest densities (from the control to about 8 plants m^{-2}) (Fig. 2). Furthermore, the values of stem diameter at 75 DAP in the control treatment, was similar to 78 plants m^{-2} at 90 DAP, evidencing the stem diameter interference due to environmental resources competition.

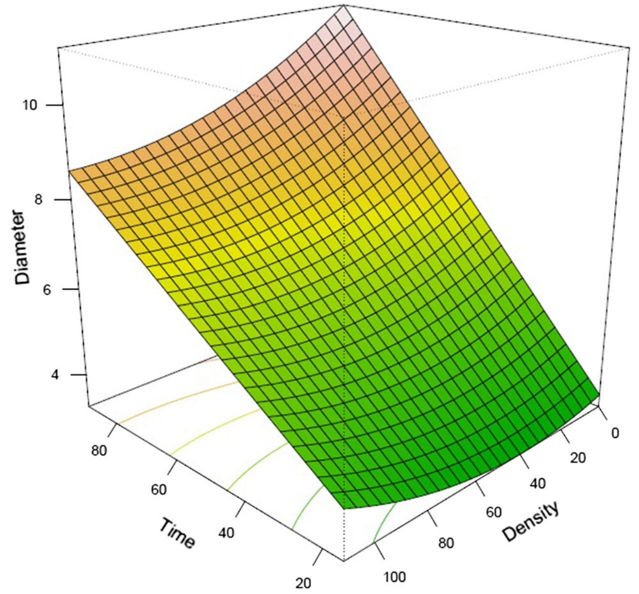

Fig. 2 Effect of increasing densities of *Urochloa brizantha* cv. Marandu on *Eucalyptus urograndis* (clone GG100) diameter (mm) during 90 days after planting. $Diameter = 1.9 + 0.1 \cdot Time - 0.009 \cdot Dens - 4 \cdot e^4 \cdot Time \times Dens - 8.8 \cdot e^6 \cdot Time^2 + 2 \cdot e^4 \cdot Dens^2$

At 90 DAP, the average diameter of the eucalyptus that coexisted in 78 plants m^{-2} or more, was 18.7% lower than the diameter in density smaller than 8 plants m^{-2}. Also, up to 12 plants m^{-2}, it was observed no reduced diameter growth greater than 5% when compared to the control (Fig. 2).

Regarding the quantum efficiency of photosystem II (PSII) (F_v/F_m), the response pattern was the same in relation to time (Fig. 3). There was a decrease in values in relation to density, where the lowest densities had the highest values, and the highest densities had lowest values.

At the end of the experimental period, the treatment with the highest density was 5.2% lower than the control. Thus, among the variables evaluated during the experiment, the F_v/F_m was the variable with less sensitive to the stress due to competition with the weed (Fig. 3).

Concerning the eucalyptus dry matter (Table 3), all plants that coexisted with *U. brizantha* showed lower values compared to the control. It should be noted that treatment with 22 plants m^{-2} also differed from the treatments with 44, 89 and 111 plants m^{-2}; and the eucalyptus plants that coexisted with the highest density of *U. brizantha* showed 63.9% less dry matter compared to control. For the leaf area (Table 3), all treatments, except that with 22 plants m^{-2}, were significantly different from control, with lower values for this variable. Treatments with 22 and 33 plants m^{-2} did not differed from each other, but showed statistical difference for the treatments with 44, 89 and 111 plants m^{-2}. Also, the treatment with the highest density of weed plant resulted in lower leaf area, being 72.7%

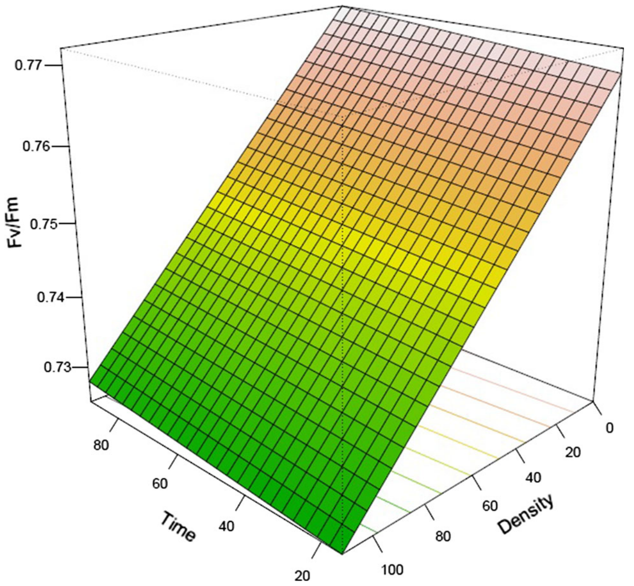

Fig. 3 Effect of increasing densities of *Urochloa brizantha* cv. Marandu on *Eucalyptus urograndis* (clone GG100) F_v/F_m during 90 days after planting. $\frac{Fv}{Fm} = 0.76 + 0.00004 \cdot Time - 0.0004 \cdot Dens$

Table 3 Average values of variables: *Eucalyptus urograndis* (clone GG100) dry matter (DMeuc—g), leaf area, net CO_2 assimilation rate (µmol m^{-2} s^{-1}), stomatal conductance (mol m^{-2} s^{-1}), transpiration rate (mmol m^{-2} s^{-1}), after 90 days of coexistence with increasing densities of *Urochloa brizantha* cv. Marandu

Densities (plants m^{-2})	DMeuc (g)	Leaf area (cm^2)	Net CO_2 assimilation rate	Stomatal conductance	Transpiration rate	DMpalisade grass (g)
0	45.05 A	3256.4 A	10.50	0.0822	1.56	–
22	33.46 B	2151.9 AB	10.15	0.1223	2.19	11.43 A
33	26.25 BC	1713.3 B	8.41	0.1091	1.96	9.78 AB
44	17.11 C	972.4 C	9.22	0.1388	2.48	8.98 AB
67	23.22 BC	1429.5 BC	12.27	0.1600	2.75	6.25 ABC
89	17.07 C	948.7 C	9.16	0.1398	2.47	5.37 BC
111	16.26 C	887.8 C	10.11	0.1508	2.63	3.89 C
F	21.13**	21.70**	1.19^{ns}	1.63^{ns}	1.90^{ns}	5.84**

Average of *U. brizantha* dry matter (DMpalisade grass—grams per plant unity)

Means followed by different letters differ from each other at the level of 5% of probability by Tukey test. **Significant at the level of 1% of probability. ^{ns}Not significant by *F*-test

lower than control, proving to be the most sensitive variable to the competition imposed by *U. brizantha*.

In the assessments of net CO_2 assimilation rate, stomatal conductance and transpiration rate of eucalyptus, held at 90 DAP (Table 3), there were no significant differences between treatments.

For *U. brizantha* dry matter (Table 3), it was observed that with increasing densities per treatment, there was a decrease of the mean values of this variable per unit of *U. brizantha*, in which the treatment with 22 plants m^{-2} differ significantly than that with 89 and 111 plants m^{-2}, and the densities of 33 and 44 plants m^{-2} had statistical difference for treatment with higher density, which showed dry matter value 65.9% lower than the treatment of lower density (22 plants m^{-2}).

Discussion

The reduced development of some eucalyptus variable, such as plants height, stem diameter, dry matter, leaf area and quantum efficiency of PSII (F_v/F_m) (Figs. 1, 2, 3; Table 3), which were observed in this study, is the result of interference imposed by *U. brizantha* plants to the eucalyptus and corroborate results obtained by many authors (Toledo et al. 2000; Adams et al. 2003; Florentine and Fox 2003; Garau et al. 2009). They elucidate that the first year after crop installation is the period in which eucalyptus are more susceptible to competition imposed by weeds. Based on a field experiment, Tarouco et al. (2009) observed that if *E. urograndis* plants were free of competition for a total period of 335 DAP, there would be no significant loss of productivity due to interference of weeds in the crop. The authors also reported that the critical period for preventing interference was from 107 to 335 DAP.

In scheme developed by Bleasdale (1960) and later adapted by Pitelli (1985), it is possible to verify that biotic and abiotic factors affect the intensity of weed community interference on crop plants, such as: soil and climatic characteristics of the region; species/clone of eucalyptus used in planting; besides species, density, and weed distribution in the field.

In this context, Bacha et al. (2016) observed that a density of 3 plants m^{-2} of *U. decumbens* leads a decreasing of 55% on leaf area and 48% on dry matter of *E. urograndis* at 90 DAP. Moreover, in the present work were required 33 plants m^{-2} of *U. brizantha* to induce losses of 47.3% on the leaf area and 41.7% on the dry matter of the same eucalyptus species. Thus, it is possible to infer that because of the different competitive capabilities that these two species of plants have, they cause physiological changes that negatively affect eucalyptus, reflecting different reduces growth in height, diameter, leaf area, among others. This result confirms the Santos et al. (2015) that suggested the outperformance of *U. decumbens* over *U. brizantha* on the interference of *E. urograndis* photosynthetic characteristics.

In this perspective, Toledo et al. (2001) observed that a density of 4 plants m^{-2} of *U. decumbens* led reductions in growth of *E. grandis* plants. Similar results were found by Dinardo et al. (2003) regarding the density of *Panicum maximum* in the same eucalyptus species, which suggests that guinea grass also presents greater competitive ability compared to *U. brizantha*.

The competition with weeds can result in low supply of some essential resources to the growth and development of eucalyptus, causing deficiencies that result in changes of some parameters related to photosynthesis, like low quality and/or amount of incident light (Sharkey and Raschke 1981) and water deficiency (Margolis and Brand 1990; Grossnickle and Folk 1993; Lamhamedi et al. 1998).

The quantum efficiency of PSII, which is represented by the fluorescence ratio F_v/F_m, is an important physiological characteristic used in studies related to different stresses (for example, the one caused by competition with weeds), because it is an indicator of the photosynthetic capacity of the plants. These values express the efficient capture of light energy by the PSII reaction center, resulting in an electron transport through this photosystem (Krause and Weis 1991).

Maxwell and Johnson (2000) emphasized that the optimal values of F_v/F_m can vary across species. Based on this study we clearly observed the F_v/F_m reduction according to weed density increasing (Fig. 3). We speculate that the low values of F_v/F_m, which was found in the treatments with the highest densities of *U. brizantha*, was the cause of the decrease in the values of the other characteristics evaluated in this study, since the photoassimilates produced on photosynthesis are used in plant growth. Also, as the gas exchange evaluations (transpiration, stomatal conductance and net CO_2 assimilation rate) did not indicate difference between the treatments (Table 3), we can infer that the limitation of the process was found in the photochemical phase of photosynthesis (indicated by the F_v/F_m ratio).

Regarding *U. brizantha* dry matter (Table 3), it is possible to observe that the larger the number of plants per unit area, smaller was the individual growth of the plants. Thus, it is evident that the competition for the environmental resources was a limiting factor for the plants growth in both occasions, to the culture (in a interspecific competition) and to the weed community (in a intraspecific competition).

Conclusion

We conclude that from the density of 22 plants m^{-2}, *Urochloa brizantha* cv. Marandu negatively interferes in the growth of *Eucalyptus urograndis* (clone GG100). Up to 8 plants m^{-2} there are no reductions greater than 5% in eucalyptus height and stem diameter.

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