Tiller population density and tillering dynamics in marandu palisade grass subjected to strategies of rotational stocking management and nitrogen fertilisation

ABSTRACT. The objective of this experiment was to evaluate tiller population density and the dynamics of the tillering process in marandu palisade grass subjected to strategies of rotational stocking management and nitrogen fertilisation. Treatments corresponded to combinations between two targets of pre-grazing conditions (sward surface height of 25 and 35 cm) and two rates of nitrogen application (50 and 200 kg ha-1 year-1), and were allocated to experimental units according to a 2x2 factorial arrangement in a randomised complete block design, with four replications. The following response variables were studied: initial (TPDi), intermediate (TPDm) and final (TPDf) tiller population density as well as the rates of tiller appearance (TAR) and death (TDR) and the tiller population stability index (SI). TPDi was similar to all treatments, with differences in tiller population density becoming more pronounced as the experiment progressed, resulting in larger TPDf on swards managed at 25 cm pre-grazing height. Tiller death was larger on swards managed at 35 cm, with differences in tiller appearance being recorded only from February 2010 onwards. Stability of tiller population was higher on swards managed at 25 cm pre-grazing height. Overall, there was no effect of nitrogen on the studied variables, and the most adequate grazing strategy corresponded to the pre-grazing height of 25 cm, regardless of the nitrogen application rate used.

KEY WORDS: Management targets; nitrogen fertiliser; tiller population; grazing management; stability of plant communities

**INTRODUCTION**

 Pastures are comprised of plant populations that, for forage grasses, tillers correspond to the basic units of growth (HODGSON, 1990). These have limited lifespan, with grazing being the most common cause of tiller death, consequence of the decapitation of growing points. In this context, it is important to ensure replacement of dead tillers as a means of assuring population stability and productivity (HERNÁNDEZ-GARAY et al., 1999). Under grazing, variations in tiller population correspond to the most effective way forage grasses have to adapt to defoliations regimes being imposed, since they allow for large flexibility in adjustments in sward leaf area index (MATTHEW et al., 2000), the main sward structural characteristic governing plant responses (LEMAIRE; CHAPMAN, 1996). Knowledge regarding the demography of the tillering process and its pattern of variation in relation to defoliation strategies used and seasons of the year favours the identification of management opportunities aiming at optimising the natural cycle of tiller appearance and death, basic condition for ensuring pasture stability and rational and sustainable use of grassland areas. Nitrogen fertiliser has an important effect on tillering, and certainly can be used in association with defoliation strategies as a means of increasing herbage production and productivity (CAMINHA et al., 2010). This highlights the importance of studies on the tillering process and plant responses to strategies of grazing and fertilisation. Against that background, the objective of this experiment was to evaluate tiller population and the dynamics of the tillering process in marandu palisade grass (*Brachiaria brizantha* ‘Marandu’) subjected to strategies of rotational stocking management and nitrogen fertilisation.

 **MATERIAL AND METHODS**

 The experiment was carried out at Nova Odessa, SP, Brazil (22o42’S, 47o18’W and 528 m a.s.l.), on a 48 ha *Brachiaria brizantha* (Hochst ex A. Rich) Stapf. cv Marandu pasture (marandu palisade grass) established in 1995 on a Rhodic Ferralsol, concomitantly to a grazing trial carried out from January 2009 to May 2010 (GIMENES et al., 2011). Average soil chemical characteristics (RAIJ et al., 1986) for the 0-20 cm layer were: pH CaCl2: 4.5; OM = 37.2 dg dm-3; P (ion-exchange resin extraction method) = 1.6 mg dm-3; Ca = 14.9 mmolc dm-3; Mg = 10.4 mmolc dm-3; K = 2.1 mmolc dm-3; H + Al = 40.7 mmolc dm-3; sum of bases = 27.4 mmolc dm-3; cation exchange capacity = 68.1 mmolc dm-3; base saturation = 39.7%, indicating low pH, phosphorus and potassium levels relative to plant nutrient requirements (WENER et al., 1996). Limestone was applied only in June 2008, after first summer, at a rate of 1.5 t ha-1 with the objective of increasing base saturation to 50%. Phosphorus and potassium were applied along with nitrogen according to experimental treatment specifications described below. Climate corresponds to the Cwa type in Köppen Classification, humid tropical with a defined rainy season during summer, dry winter and 22 oC of average annual air temperature and rainfall around 1200 mm. At the experimental site, the average annual rainfall is 1270 mm, with around 30% concentrated from May to September. Further details on climatic conditions during the experimental period were presented by (GIMENES et al ., 2011)

 Pasture was originally divided into 48 paddocks of approximately 1 ha each, which were being laxly grazed under continuous stocking since 2006 and presented high herbage masses comprised of large proportions of stem and dead material. Prior to the beginning of the experiment, four blocks of 12 paddocks each were identified and preparation of the area started in September 2008 with partial removal of herbage mass by grazing one block at a time. As grazing was completed, paddocks were mowed at 20 cm, a procedure that started in October and finished in December 2008. Paddocks were further divided into two halves, increasing the number of paddocks per block to 24 and the total number of paddocks to 96 (approximately 0.5 ha each). Treatments corresponded to combinations between two pre-grazing heights (25 and 35 cm, equivalent to 95% and maximum canopy light interception - LI, respectively (TRINDADE et al., 2007; GIACOMINI et al., 2009a,b) and two levels of nitrogen application (50 and 200 kg ha-1 year-1, equivalent to maintenance (CADISH et al.,1994; WERNER et al.,1996) and production (FAGUNDES et al., 2005; OLIVEIRA et al., 2005) levels, respectively) (designated as 25/50, 25/200, 35/50 and 35/200 from this point onwards), and were allocated to experimental units according to a randomised complete block design, with four replications. Each experimental unit was comprised of six 0.5 ha paddocks (totalling 3 ha), which were managed as a farmlet. Each farmlet received three Nellore steers (average initial body weight of 330 kg) as test animals for measuring weight gain plus a variable number of extra steers to adjust stocking rate and allow grazing to be executed according to management specifications for individual treatments. Animal shrunk weight (12 hours) was recorded every four weeks, and data used to adjust stocking rate and calculate animal weight gain per unit area. Target post-grazing height was set at 15 cm; although this was not a fixed value since at times rate of rotation had to be increased to cope with fast pasture growth. The main focus was to keep pre-grazing conditions on target (a variation of only 5% around the target was allowed throughout the experiment) by using stocking rate and rotation length (by means of manipulating the duration of the occupation period of paddocks) as management decisions. Priority was given to adjustments in stocking rate when animal numbers were sufficient, but, when pasture growth was larger than demand and not enough animals were available, rotation was accelerated by shortening occupation period and leaving a post-grazing residue taller than 15 cm. In order to execute that, farm walks were performed in a weekly basis and decisions made taking into account the condition of the paddock being currently grazed and that of the following two in the regrowth sequence. Extra animals were maintained in a 20 ha marandu palisade grass pasture adjacent to the experimental area. A total of 420 steers were used in the experiment.

 Nitrogen was applied in instalments throughout summer (January to March, 2009 and 2010) along with phosphorus and potassium (Table 1). Monitoring of experimental conditions was made through systematic readings of sward surface height using a sward stick (BARTHRAM, 1985) along pre-defined zig-zag transect lines covering the entire area of each paddock at pre- and post-grazing (100 readings per paddock). Measurements of sward height during regrowth were made in a weekly basis. Light interception by sward canopy was also measured every two months with the objective of checking if the pre-grazing height targets of 25 and 35 cm were consistent with the 95 and 100% LI conditions, respectively. Measurements were carried out on 6 randomly selected locations from areas representative of sward condition at the time of sampling (visual assessment of herbage mass and height) using a canopy analyser (LAI 2000, LI-COR, Lincoln, Nebraska, USA) (DA SILVA et al., 2009). In each location, 1 reading was taken above the canopy and 5 at ground level, totalling 6 readings above the canopy and 30 at ground level per paddock.

**Table 1 –** Times and rates of fertiliser application on marandu palisade grass subjected to strategies of rotational stocking management from January 2009 to April 2010

|  |  |  |
| --- | --- | --- |
|  | Nitrogen application rate (kg ha-1) |  |
| Month | 50 | 200 | Fertiliser |
| January | 50 | 50 | 20:2:17\* |
| February | 0 | 75 | Urea |
| March | 0 | 75 | Urea |

 \* Commercial N:P:K formula

 Evaluations of tiller population and tillering dynamics started only in late October 2009 (mid-spring), since the first summer and autumn (January to June 2009) were considered as a period of adaptation of swards to grazing treatments imposed in the grazing trial. Measurements were made in pre-defined sampling paddocks, one per farmlet, chosen at the beginning of the grazing experiment and used consistently throughout the experimental period. In each sampling paddock, after the first grazing in spring, three sampling areas, representative of the average sward condition (visual assessment of herbage mass and height), were marked with a 30 cm-diameter PVC ring fixed on the ground by metallic staples and all tillers within them were counted and identified using white plastic coated wire. Following that, after every grazing, live and dead tillers within the rings were counted and new tillers tagged using a different colour each time. Tillers were considered dead when they were dry or in an advanced stage of senescence, or had disappeared. The procedure allowed tillers from all generations formed to be counted and new ones recorded and data was used to calculate the rates of tiller appearance (TAR), survival (TSR) and death (TDR) (tiller.tiller-1.day-1).

Measurements of tiller population density (TPD) were carried out independently from those of tillering dynamics using three 1.00 x 0.25 m metallic frames per sampling paddock. These were randomly placed on representative areas of the paddocks at the time of sampling (visual assessment of herbage mass and height). The procedure was executed before every grazing and data was grouped in three periods: initial (TPDi), intermediate (TPDm) and final (TPDf). TPDi was calculated using data from the first measurement only, which was performed from October to December 2009. TPDm was calculated with data recorded from January to March 2010, and TPDf was calculated using data from the last measurement only, which was performed from March to May 2010.

Analysis of variance was performed using the Mixed Procedure of SAS® (Statistical Analysis System). The choice of the covariance matrix was made using the Akaike Information Criterion (AIC) (WOLFINGER, 1993), and analysis performed considering pre-grazing height, nitrogen application rate, month and/or period of the experiment for the TPD data and their interactions as fixed effects and blocks as a random effect (LITTEL et al., 2000). When appropriate, treatment means were calculated using the “LSMEANS” statement, and comparisons made with “PDIFF” based on a Student t test and a 5% significance level.

**RESULTS AND DISCUSSION**

 Tiller population density (TPD) varied only with period of the experiment (P<0.0001) and pre-grazing height x period of the experiment interaction (P=0.0220). Overall, TPD increased from the beginning to the end of the experimental period, with differences between pre-grazing heights becoming significant only at the end of the experiment, when larger values were recorded on swards managed with the 25 relative to the 35 cm pre-grazing height (Table 2).

# Table 2 – Tiller population density (tillers m-2) on marandu palisade grass swards subjected to strategies of rotational stocking management characterised by the pre-grazing heights of 25 and 35 cm from October 2009 to April 2010

|  |  |  |
| --- | --- | --- |
| Period of the experiment | Pre-grazing height (cm) | SEM |
| 25 | 35 |
| Initial | 765 Ab | 864 Ab | 57.8 |
| Intermediate | 1302 Aa | 1185 Aa | 57.8 |
| Final | 1471 Aa | 1227 Ba | 57.8 |

Means followed by the same upper case letters in lines and lower case letters in columns are not different (P>0.05). SEM = Standard error of the mean.

 Such pattern of response is associated with variations in climatic conditions throughout the experiment (GIMENES et al., 2011), the magnitude of the differences being determined by the grazing treatment imposed. According to Langer (1963), variations in climatic conditions related to seasons of the year interfere with the patterns of tillering, since the process is strongly influenced by the availability of light, water, temperature and nutrients, particularly nitrogen.

 Swards managed with the pre-grazing height of 25 cm were grazed more frequently, with shorter occupation periods and smaller herbage mass than those managed with the pre-grazing height of 35 cm. This resulted in larger number of grazing cycles (GIMENES et al., 2011), generating better conditions for tillering related to a more appropriate light environment. Swards managed with longer grazing intervals and greater pre-grazing height and larger herbage mass, as those managed at 35 cm, usually show lower TPD than swards defoliated more frequently, with lower pre-grazing height and herbage mass (MATTHEW et al., 2000), that being related with the quality of the light environment within the canopy (GAUTIER, 1999). Light is a critical resource for forage grasses, since growth is determined by the interception of photosynthetically active radiation (PAR – 400 to 700 nm) and its distribution within the sward canopy (PEARSON; ISON, 1997). The fact indicates that both quantity and quality of the light within sward canopy may have been determinant of the variation in TPD on swards managed with the pre-grazing height of 35 cm.

 Under rotational stocking management conditions, tropical forage grasses show reduction in leaf elongation and increase in stem elongation and senescence after 95% of sward light interception during regrowth (DA SILVA; NASCIMENTO JUNIOR, 2007), clearly indicating significant competition for light under those circumstances, interfering with the processes of tiller appearance and death and, as a result, with tiller population density of swards (HERNÁNDEZ-GARAY et al., 1999). For marandu palisade grass such condition of 95% LI corresponds to the pre-grazing height of 25 cm (TRINDADE et al., 2007), indicating that variations in TPD recorded in large scale experiments (grazing trials) follow the same pattern of variation described and measured in smaller scale experiments (paddocks) (GIACOMINI et al., 2009a; SBRISSIA; DA SILVA, 2008).

 Tiller appearance rate (TAR) was influenced by pre-grazing height (P=0.0242), month of the year (P=0.0001) and pre-grazing height x month of the year interaction (P=0.0011). There was no difference in TAR between swards managed at 25 and 35 cm pre-grazing height in December 2009 and January 2010, with larger values recorded on those managed at 35 cm in February and March 2010 (Table 3). In general, tiller appearance decreased as the experiment progressed, with larger values recorded in December 2009 and January 2010, and lower values recorded in March 2010. On swards managed at 25 cm pre-grazing height, the decrease in tiller appearance started in February 2010 and on those managed at 35 cm in March 2010 only. On the other hand, tiller death remained relatively stable throughout the experimental period, with larger values recorded on swards managed at 35 relative to those managed at 25 cm pre-grazing height, the difference varying from 45 to 67% from December 2009 to March 2010 on swards fertilised with 50 kg ha-1 of N and decreasing from 110 in December 2009 to 7% in March 2010 on those fertilised with 200 kg ha-1 (Table 4).

# Table 3 – Tiller appearance rate (tiller tiller-1 day-1) on marandu palisade grass subjected to strategies of rotational stocking management characterised by the pré-grazing heights of 25 and 35 cm from December 2009 to March 2010

|  |  |  |
| --- | --- | --- |
| Month | Pre-grazing height (cm) | SEM |
| 25 | 35 |
| December/09 | 0.43 Aa | 0.45 Aa | 0.033 |
| January/10 | 0.40 Aa | 0.47 Aa | 0.033 |
| February/10 | 0.28 Bb | 0.48 Aa | 0.033 |
| March/10 | 0.23 Bc | 0.36 Ab | 0.033 |

Means followed by the same upper case letters in lines and lower case letters in columns are not different (P>0.05). SEM = Standard error of the mean.

# Table 4 – Tiller death rate (tiller tiller-1 day-1) on marandu palisade grass subjected to strategies of rotational stocking management and fertilised with nitrogen from December 2009 to March 2010

|  |  |
| --- | --- |
|  | Rate of N fertilisation (kg ha-1) |
| Pre-grazing height (cm) | 50 | 200 |
|  | *December/09 (SEM=0.037)* |
| 25 | 0.27B | 0.20 B |
| 35 | 0.40 A | 0.42A |
|  | *January/10 (SEM=0.037)* |
| 25 | 0.29B | 0.23 B |
| 35 | 0.42 A | 0.42 A |
|  | *February/10 (SEM=0.037)* |
| 25 | 0.24 B | 0.25 B |
| 35 | 0.40 A | 0.44A |
|  | *March/10 (SEM=0.037)* |
| 25 | 0.25 B | 0.29 B |
| 35 | 0.41 A | 0.31 AB |

Treatment means (rate of N application/pre-grazing height) within months of the year followed by the same upper case letter are not different (P>0.05). SEM = Standard error of the mean.

The increase in tiller numbers occurs, primarily, as a means of decreasing the distance between plants and increasing the occupation of the area before contributing to an increase in sward height (HERNÁNDEZ-GARAY et al., 1999). In the present study that was accomplished mainly by the production of small tillers (GIACOMINI et al., 2009a) with growing points positioned below the post-grazing height on swards managed at 25 relative to those managed at 35 cm pre-grazing height. Only in March 2010, time of reproductive growth and flowering, the reduction in tiller appearance was smaller on swards subjected to the 35/200 treatment than on those subjected to the 35/50 treatment, a condition likely associated with larger proportion of aerial tillers favoured by the decapitation of reproductive meristems and larger post-grazing heights. Further, aerial tillers are positioned closer to the stems of parent tillers, making decapitation of growing points difficult to happen. Such condition may have resulted in the lower mortality of tillers recorded for the 35/200 treatment at that time of the year. Probably, if the measurement period had been longer, similar pattern of response would have been observed for treatment 35/50, since the same sward conditions and structural characteristics were generated relative to the 35/200 treatment, the only difference being the timing of decapitation of the reproductive growing points, consequence of the N application rates used.

Tiller appearance remained relatively stable during the first two months of the experimental period (December 2009 and January 2010), with differences between pre-grazing heights becoming significant from February 2010 onwards (Table 3). Decrease in tiller appearance started initially on swards managed at 25 cm, in February 2010, when recorded values of TAR were 42% lower than those recorded on swards managed at 35 cm. In March 2010 tiller appearance continued to decrease on swards managed at 25 cm and also on those managed at 35 cm (reduction from 17.8 and 25.0% for 25 and 35 cm relative to February, respectively). Reproductive growth of the marandu palisade grass usually starts in February and reaches its peak in March (CAMINHA et al., 2010), suggesting decapitation of reproductive meristems soon at the beginning of the reproductive development, in February, on swards managed at 25 cm relative to those managed at 35 cm pre-grazing height, consequence of the higher frequency of defoliation under those circumstances. Again, the main aspect here would be the timing of defoliation of reproductive tillers that, in this case, was basically a function of the grazing interval generated by the targets of pre-grazing height used. In the case of early decapitation on swards managed at 25 cm, the production of aerial tillers is low since stem elongation is small (GIACOMINI et al., 2010). That, in association with the reduction in availability of climatic growth factors (GIMENES et al., 2011), generated inadequate conditions for aerial tillering and explains the continuous reduction in tiller appearance until the end of the experiment. In the case of late decapitation of reproductive tillers on swards managed at 35 cm, there was the presence of decapitated stems serving as substrate for the production of aerial tillers that, in spite of being smaller and shorter lived relative to basal tillers (GIACOMINI et al., 2009a), contributed to decrease the impact of the reduction in tiller appearance under those circumstances.

 As a result of the drastic reduction in tiller mortality associated with the 35/200 treatment at the end of the experiment, in March 2010 there was a contrast between treatment 35/50 and those of pre-grazing height 25 cm (25/50 and 25/200). At the beginning of every new pasture growing season the tillering process is intensified, mainly through the formation of basal tillers (GIACOMONI et al., 2009a; SBRISSIA et al., 2010). Basal tillers show significant stem elongation when swards are managed under rotational stocking using maximum LI as pre-grazing target (GIACOMINI et al., 2009a), a condition that favours the decapitation of growing points and results in higher tiller mortality relative to the 95% LI pre-grazing target. Further, since the decapitated stems are substrate for the formation of aerial tillers, this category of tillers is increased significantly on swards managed at maximum LI, particularly during the phase of reproductive growth of plants (GIACOMINI et al., 2009a; SBRISSIA et al., 2010). The lower values of leaf-to-stem ratio in the herbage mass post-grazing of swards managed at 35 cm pre-grazing height, in spite of the larger values of height and herbage mass (GIMENES et al., 2011), are indicative of a smaller residual leaf area index and/or more severe defoliation and greater availability of decapitated stems, favouring the increase in the population of aerial tillers. The fact, associated with initially low values of tiller population density (765 and 864 tillers m-2 on swards managed at 25 and 35 cm, respectively – Table 2), explains the lower mortality of tillers on swards managed at 25 cm pre-grazing height, since stem elongation under those grazing conditions is low.

The tiller population stability index (Pf/Pi) varied with pre-grazing height (P=0.0059) and with the pre-grazing height x nitrogen application rate x month of the year interaction (P=0.0057). Overall, swards managed at 25 cm pre-grazing showed stability index close to 1, which were higher than those recorded on swards managed at 35 cm (Table 5), particularly for the 200 kg ha-1 nitrogen application rate from December 2009 to February 2010 and for the 50 kg ha-1 rate in March 2010.

Table 5 – Tiller population stability index (Pf/Pi) on marandu palisade grass subjected to strategies of rotational stocking management and fertilised with nitrogen from December 2009 to March 2010

|  |  |
| --- | --- |
| Pre-grazing height (cm) | Rate of N fertilisation (kg ha-1) |
| 50 | 200 |
|  | *December/09 (SEM= 0.049)* |
| 25 | 1.00 AB | 1.13 A |
| 35 | 0.87 BC | 0.82 C |
|  | *January/10 (SEM= 0.049)* |
| 25 | 0.97 AB | 1.09 A |
| 35 | 0.86 BC | 0.83 C |
|  | *February/10 (SEM= 0.049)* |
| 25 | 0.99 A | 0.93 AB |
| 35 | 0.88 AB | 0.82 B |
|  | *Março/10 (EPM= 0.049)* |
| 25 | 0.95 A | 0.84 AB |
| 35 | 0.79 B | 0.95 A |

Treatment means (rate of N application/pre-grazing height) within months of the year followed by the same upper case letter are not different (P>0.05). SEM = Standard error of the mean.

The combined analysis of the tiller appearance and death data, illustrated by the values of Pf/Pi above, indicate that tiller appearance was relatively more important than tiller death in determining the demographic patterns of tiller population, since that was the most responsive variable to treatments imposed, determining the balance between tiller appearance and death and the stability of tiller population (Table 5). In general, the increase in tiller population density was larger on swards managed at 25 cm relative to those managed at 35 cm during December 2009 and January 2010, consequence of a larger balance between tiller appearance and death and values of Pf/Pi larger than 1, particularly for the 200 kg ha-1 rate of N application (Table 5).

On swards managed at 35 cm pre-grazing the balance between tiller appearance and death was positive, although near to zero, indicating smaller increases in tiller population density relative to swards managed at 25 cm (Table 2). From February 2010 onwards, the balance near zero between tiller appearance and death indicates stabilisation of tiller population density on swards subjected to the 25/200 treatment. The fact did not happen on swards subjected to the 35/50 treatment, probably because of the slower growth and low tillering rate, highlighting, once again, the effect of treatments interfering with the timing only and not altering the pattern of response itself. In March 2010 the balance between tiller appearance and death became close to zero and/or negative, indicating the beginning of tiller mortality dependent of tiller population (intra-specific competition), since availability of climatic growth factors were starting to become limiting and, as a result, swards have to decrease tiller population as a means of adapting to those conditions during the following dry period (autumn, winter and early spring), completing the annual cycle of variation with low values of tiller population at the end of the next spring. Exception to that pattern was the treatment 35/200, whose balance between tiller appearance and death remained positive, probably because of the greater production of aerial tillers as discussed above. Such results corroborate those of the stability index (Table 5), which indicated the same pattern of response.

**CONCLUSIONS**

* Measurements of tiller population density and tillering dynamics in large scale experiments (grazing trials) provide important additional and complementary information to those relative to herbage production and animal performance for defining grazing management strategies, since they provide information on the stability of tiller population and contribute to reduce the risk of degradation of pastures.
* The most adequate grazing strategy corresponded to that characterised by the pre-grazing height of 25 cm, regardless of the nitrogen application rate used, highlighting the importance of efficient harvest of the produced herbage before considering the use of nitrogen fertiliser.

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