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Reducing supplementation frequency for Nellore beef steers grazing tropical pastures

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ABSTRACT: Reduced supplementation frequency is a broadly applied management practice. Ruminants consuming low quality forages/pastures, supplemented less than once daily are able to maintain body weight gain (BWG), efficiency of use of dry matter, nitrogen and other nutrients, as compared with animals supplemented once daily. We evaluated the feeding behavior, dry matter intake (DMI), dry matter and organic matter digestibility (DMD and OMD), BWG, *Longissimus* muscle area and backfat depth of Nellore steers raised on *Brachiaria brizantha* cv. Marandu pastures during the dry season, with different supplementation patterns. Thirty six animals (338 ± 40.7 kg) were distributed over nine paddocks according to a completely randomized design. Treatments were based on supplementation frequency: once daily (OD), once daily except Saturdays and Sundays (SS), or on alternate days (AD), at 1.0 %, 1.4 % and 2.0 % BW, respectively. Average total DMI accounted for 1.6 % BW day^{-1}, with no effect of supplementation frequency. Supplementation frequency had no effect on BWG or grazing time during the day. There was no difference in *Longissimus* muscle area animals supplemented daily, SS and AD. The backfat depth was thinner in animals supplemented AD, but even in this case, it was within the standards considered satisfactory for a finishing steer. Reducing supplementation frequency seems a good option to lower labor costs without affecting feed efficiency or carcass quality in beef cattle grazing tropical pastures.

Introduction

Forage intake is the most influential factor when assessing animal performance in grazing animals, and is affected by animal, pasture and weather characteristics, as well as by their interactions (Black and Griffiths, 1975; Webster, 1992). In tropical and subtropical areas, the availability and quality (defined by digestibility and crude protein (CP) content) of pastures for beef cattle is subject to great variations throughout the year. These variations are greater than those occurring in temperate areas, where production is a more important constraint than quality.

Reduced supplementation frequency is a broadly applied management practice. Beaty et al. (1994), Farmer et al. (2004) and Currier et al. (2004), demonstrated that ruminants consuming low quality forages/pastures, and supplemented less than once daily, are able to maintain BWG, efficiency of DM use, nitrogen and other nutrients, and efficiency of microbial synthesis, as compared with animals supplemented once daily. However, studies dealing with the effects of reduced supplementation frequency in cattle grazing tropical pastures are scarce, as a result of the number and complexity of variables to be taken into account under the conditions prevailing.

Supplementation is then likely to affect intake and digestibility of the basal diet (pasture) in a substitutive, additive or mixed way (Moore, 1980). A substitution effect is expected when infrequent supplementation is provided to cattle grazing low-quality, tropical forages during the dry season, probably as a result of the greater amount of supplement provided on supplementation days as compared with more frequent supplementation rates. Feeding behavior may also be affected by the composition, amount and pattern of administration of the concentrate, which in turn affects the amount of supplement and pasture consumed, and subsequent animal performance.

This study aimed to assess the effect of supplementation frequencies on feeding behavior, DM intake and digestibility, average BWG and carcass quality (*Longissimus* muscle area and backfat depth), in Nellore beef steers grazing *Brachiaria brizantha* cv. Marandu during the dry season in Brazil.

Materials and Methods

Experimental animals, area and treatments

Thirty six Nellore castrated steers, two years old and with an average body weight (BW) of 338 ± 40.7 kg at the beginning of the experiment, were distributed over nine paddocks (2 ha each) of *Brachiaria brizantha* cv. Marandu (four animals per paddock). Of these 36 Nellore steers, nine animals were fitted with ruminal and duodenal cannulas (one animal per paddock). Treatments were based on supplementation frequency, with supplement fed (at 08h00 in troughs placed in the paddocks) once daily (OD), once daily except Saturdays and Sundays (SS), or on alternate days (AD), at 1.0 %, 1.4 % and 2.0 % BW, respectively (equivalent to 1 % BW day^{-1}, as-fed basis, for all three treatments). Three paddocks were assigned to each treatment. Steers were placed in the paddocks two weeks before the start of the experiment, and adaptation to the supplement began at the same time. Animals remained permanently in the paddocks for the whole length of the experiment, which lasted from July to Nov 2006, totaling 144 d.

Initially, the supplement was formulated to have a minimum of 12 % CP in the dietary DM for finishing cattle (NRC, 1984), assuming the low CP of the available forage during the experimental period. Thus, the supplement was formulated to contain 21 % CP in July (18 % cottonseed meal, 79.5 % citrus pulp and 2.5 % urea (44.8 g N kg^{-1} DM) on a DM basis), 25 % CP in Aug and Sept (27.5 % cottonseed meal, 70 % citrus pulp and 2.5 % urea), and 28 % CP in Oct and Nov (35.5 % cottonseed meal, 62 % citrus pulp and 2.5 % urea. Chemical composition of cottonseed meal and citrus pulp is shown in Table 1.

Data collection and analytical procedures

Biomass availability and pasture quality were assessed monthly from July to Nov, corresponding to d 2, d 43, d 79, d 112 and d 143 of the experimental period. Forage production was measured by randomly throwing five 1 m × 1 m metallic squares into each paddock and cutting forage inside to ground level. Half of the collected biomass was separated into green leaves, stems and dead material, and the rest was used for chemical analysis, including n-alkanes concentration.

Due to the cost and difficulty of performing the technique adopted (n-alkanes) with Nellore animals, twenty-seven animals were chosen for estimating total dry matter intake, nine less-reactive animals from each treatment (three animals from each paddock; two animals without cannula and one cannulated). Forage and supplement intake, and digestibility of the whole diet, were measured over two 12-day periods (Aug and Sept) using the n-alkanes technique. The first 7 d of each period (d 35-41 in Aug and d 70-76 in Sept of the experimental period) were designated for stabilization of the alkanes in the digesta. On d 8 to 12 of the alkane dosing, fecal samples were simultaneously collected twice-daily (at 7h00 and 17h00) in Aug (d 42-46 of the experimental period) and Sept (d 77-81 of the experimental period), directly from the rectum of each animal, and frozen at -20 °C for analysis of n-alkanes concentration. At the end of each period, composite samples were formed for each animal based on the dry mass of the samples.

Table 1 – Chemical composition of the citrus pulp and the cottonseed meal used in the supplement.

	Ingredients	
	Citrus pulp	Cottonseed meal
DM, g kg^{-1}	896	907
Ash, g kg^{-1} DM	68	67
CP, g kg^{-1} DM	70	476
Ether extract, g kg^{-1} DM	25	14
NDF, g kg^{-1} DM	222	340
ADF, g kg^{-1} DM	168	205

DM: dry matter; CP: crude protein; NDF: Neutral Detergent Fiber; ADF: Acid Detergent Fiber.

Samples of leaves, stems and dead material from Brachiaria, supplement ingredients and feces were milled through a 1 mm sieve. After this, 0.5 g was weighed and placed into 200 × 20 mm thick-walled screw-topped Pyrex test-tubes, adding 100 mg of internal standard (solution of heptane containing 1 mg g^{-1} of C_{22} and C_{34}). Then n-alkanes were extracted following the technique described by Mayes et al. (1986), with the modifications suggested by Keli et al. (2008).

Alkane analysis was carried out by on-column injection of 0.2 µL of the eluate onto a 30 m × 0.530 mm HP-1 capillary column (1.5 µm thickness) in an Agilent 6890 gas chromatograph fitted with an automatic injector and flame ionization detector. The carrier gas was helium (10 mL min^{-1}) as was the make-up gas to the detector (45 mL min^{-1}). The injector was programmed to track the oven's temperature program which was as follows: 230 °C for 0.2 min and a ramp of 6 °C min^{-1} to 300 °C, maintained for a further 18 min. Equilibrium time was set at 5 min. The detector was maintained at 350 °C throughout the whole process. Peak area data were processed using the HP ChemStation software (version A.08.03). Detector response factors for individual n-alkanes were determined by injecting onto the chromatograph a standard n-alkane mixture (C_{21}–C_{36} inclusive) after every eight sample extracts.

The intake of forage components (leaves, stems and dead material) and concentrate was estimated using the EatWhat program developed by Dove and Moore (1996). This requires the correction of alkane profiles of dietary components for fecal recovery, and for this purpose the values obtained by Morais et al. (2011) in a parallel experiment with the same animals fed palisade grass indoors were used.

Digestibility of DM (DMD) and OM (OMD) was calculated using the natural alkane C_{31} as an internal marker as follows (Mayes et al. 1986):

$$DMD/OMD = 1 - \frac{I_{31}}{F_{31}}$$

where I_{31} and F_{31} are the concentrations of C_{31} in intake and feces (recovery-corrected), respectively. The former was estimated from the calculated proportions of Brachiaria leaves, stems and dead material, and supplement in the diet. Concentration of C_{31} was expressed as mg kg^{-1} DM or mg kg^{-1} OM in order to obtain DMD or OMD, respectively.

Feeding behavior was studied over three consecutive days in Aug (d 24-26 of experimental period) in all animals. During these three days animals were receiving supplementation, and visual observations were taken once every 15 min from 06h00 to 18h00. Trained observers (three per paddock, working shifts of four hours each) recorded the grazing/non-grazing activity of the four animals in each paddock at the same time. Animals in SS and AD treatments were observed during the first day, those in OD and SS treatments during the second day, and those in OD and AD treatments during the third day.

To ascertain performance and carcass characteristics, 27 Nellore steers (without cannula) were chosen, selecting three animals per paddock. Body weight was recorded at the beginning of the experiment (d 0) and every four weeks afterwards (d 29, d 57, d 86, d 115, and d 143 of the experimental period). The weight measurements were taken in the morning, before supplementation, without solid and liquid fasting. At the same time, real-time ultrasonographic measurements (Piemedical Aquila equipment fitted with a 3.5 MHz echo sounder) of *Longissimus* muscle area and backfat depth (both between 12th and 13th ribs) were taken (Silva et al., 2005).

The N (AOAC Official Method 984.13) and ash (AOAC Official Method 942.05) concentrations in pasture (whole, leaves, stems and dead material), supplement components (cottonseed meal and citrus pulp) and feces (ashes only) samples were determined in accordance with AOAC (1995). The supplement ingredients were also analyzed for ether extract (EE) as described by AOAC (1995; Official Method 920.39). The concentrations of neutral detergent fiber (NDF) and acid detergent fiber (ADF) in pasture and supplement components were determined using the method proposed by Van Soest and Robertson (1985), with samples being digested for 40 min at 110 °C and 50.7 kPa in an autoclave (Senger et al., 2008). Acid detergent lignin (ADL) was determined according to Goering and Van Soest (1970).

Experimental design and statistical analysis

A completely randomized design was employed with three treatments (supplementation frequency) and three replications (paddocks). Herbage allowance and quality (chemical composition), dry matter intake (DMI), digestibility (DM and OM), body weight gain animal (BWG), carcass characteristics (LMA and BD) and grazing time data were subjected to an analysis of variance as repeated measures, using the PROC MIXED in SAS (Statistical Analysis System, version 9.0). Data were analyzed as a mixed model with the fixed effects of supplementation frequency, month of study and their interaction, and random residual error. Various variance and covariance structures of errors were tested for each variable. The selection of the structure was based on the lowest Bayesian information criterion (BIC).

The covariance structures FA(1) (DMI pasture, kg day^{-1}); CS (DMI pasture, kg 100 kg^{-1} BW and DMI supplement, kg day^{-1}); UN(1) (DMI supplement, kg 100 kg^{-1} BW and DMI total, kg day^{-1} or kg 100 kg^{-1} BW); VC (DM and OM digestibility, and BWG) and CSH (LMA and BD), were used. Time around which grazing activity concentrated was analyzed using Rayleigh's uniformity test of the Oriana (Kovach Computing Services, version 2.02). Mean separation between mean values were tested using Tukey's test, and significance was declared at $p \leq 0.05$. Initial live weight was used as a covariate when analyzing BWG and carcass characteristics data.

Results and Discussion

Biomass availability and composition

Biomass availabilities per ha (total, green leaves, stems and dead material) and per 100 kg BW (total and green leaves), during the months studied, are shown in Table 2. Differences between months ($p < 0.05$) were observed for all variables studied. Total biomass availability was, on average, 4.5 t DM ha^{-1}, with the highest ($p < 0.05$) values in July (5.9 t DM ha^{-1}). On average, 18% of the available biomass was green leaves, 59% stems and 24% dead material. These values, however, changed between months, with the forage collected in Oct and Nov having more leaves (31% and 32%, respectively),

Table 2 – Initial body weight animals (BW), biomass availability and chemical composition of *Brachiaria brizantha* cv. Marandu (n = 9).

	Months					SEM	*p*
	July	Aug	Sept	Oct	Nov		
BW, kg	348.9 b	357.2 b	361.3 b	367.3 ab	392.3 a	7.26	< 0.001
Total biomass[1]	5.9 a	3.8 c	3.8 c	4.1 bc	4.8 b	0.22	< 0.001
Total biomass[2]	30.2 a	18.9 b	18.8 b	19.9 b	21.7 b	1.14	< 0.001
Green leaves, %	8.6 b	6.6 b	9.0 b	31.2 a	32.4 a	0.84	< 0.001
Green leaves [2]	2.6 b	1.2 c	1.7 bc	6.3 a	7.0 a	0.23	< 0.001
Stems, %	59.5 b	69.3 a	64.8 ab	49.8 c	50.1c	1.26	< 0.001
Dead material, %	31.9 a	24.1b	26.1 b	19.0 c	17.5 c	1.08	< 0.001
	Chemical composition						
DM, g kg^{-1}	644 a	694 a	641 a	426 b	438 b	1.7	< 0.001
Ash, g kg^{-1}DM	62 b	60 b	61 b	71 a	73 a	0.2	< 0.001
CP, g kg^{-1}DM	30 b	27 b	30 b	48 a	43 a	0.2	< 0.001
NDF, g kg^{-1}DM	774 a	789 a	752 a	742 ab	696 b	1.4	< 0.001
ADF, g kg^{-1}DM	461 a	470 a	460 a	427 ab	403 b	1.1	< 0.001
ADL, g kg^{-1}DM	77	76	82	75	69	0.3	0.068

[1]t DM ha^{-1}; [2]kg 100 kg^{-1} BW; DM: dry matter; CP: crude protein; NDF: Neutral Detergent Fiber; ADF: Acid Detergent Fiber; ADL: Acid Detergent Lignin; SEM: Standard error of the mean; p: Probability of the differences; a, b,c: Means followed by different letters are different ($p < 0.05$).

and less stems (50%) and dead material (19 % and 17%), than the pasture harvested in July (9% green leaves, 59% stems and 32% dead material, respectively), Aug (7%, 69% and 24%) or Sept (9%, 65% and 26%). These last two months showed the lowest biomass availability (3.8 t DM ha^{-1}) together with the highest proportions of stems in it (67 % on average). The high biomass availability observed in July (5.9 t DM ha^{-1}) matched with the highest proportion of dead material (32%).

Herbage allowance (kg 100 kg^{-1} BW) was also higher ($p < 0.05$) in July, as a consequence of the greater biomass production and the lower weight of the animals. On the other hand, offer of green leaves (kg DM 100 kg^{-1} BW) was also higher in Oct and Nov ($p < 0.05$). As expected, stocking rate (not shown), calculated as kg BW 450^{-1} (weight of the standard animal) per ha, increased from July (1.55) to Nov (1.74), although differences were significant only for this last month.

Table 2 shows the chemical composition of available pasture from July through Nov. All analyzed parameters, except lignin ($p > 0.05$), were influenced by the month studied ($p < 0.05$). Crude protein content increased in Oct and Nov, whereas DM, NDF and ADF decreased, probably as a result of higher rainfall (Oct = 184.5 mm; Nov = 166.8 mm) than in other months (July = 3.2mm; Aug = 19.1 mm; Sept = 37.6 mm), which led to pasture regrowth with a higher proportion of green leaves, and hence better quality.

The n-alkanes concentration in green leaves, stems and dead material from *Brachiaria brizantha* cv. Marandu, and the concentrate used in the present experiment is shown in Table 3. Only data for Aug and Sept are given as these were the two months when intake studies were performed. Leaves had the highest concentration in total alkanes (332.4 mg kg^{-1} DM on average), with a predominance of C_{31} and C_{33}. These two hydrocarbons were also the most abundant in the dead material and the stems, with an average concentration in total alkanes of 180.6 mg kg^{-1} DM and 91.7 mg kg^{-1} DM, respectively. As expected, the concentrate showed the lowest content in paraffins (53.9 mg kg^{-1} DM), and in these case C_{25} and C_{23} were the alkanes in higher proportions. The observed marked decrease in concentrations from Aug to Sept for green leaves is in accordance with the decline (although not significant) in NDF content of the *Brachiaria* as a whole (Table 2). The chemical composition of stems (1.2 % vs. 1.6 % CP, and 81.2 % vs. 81.7 % NDF for Aug and Sept, respectively) and dead material (2.3 % vs. 2.5 % CP, and 75.1 % vs. 74.7 % NDF) changed to a much lesser extent than that of leaves (8 % vs. 14.4 % CP, and 63.2 % vs. 61.4 % NDF), and hence the decline in alkane concentrations was much lower (stems) or did not exist at all (dead material).

Intake, diet composition and digestibility

Supplementation frequency did not affect total, pasture or DM concentrate intake (Table 4), neither as kg day^{-1} nor as kg 100 kg^{-1} BW ($p > 0.05$). On the other hand, the month of study had an effect on all variables ($p < 0.05$) except total and DM concentrate intake expressed as kg 100 kg^{-1} BW ($p > 0.05$). The interaction between supplementation frequency and month of study was not significant in any case ($p > 0.05$).

Intake studies using alkanes were also tried in Nov. However, estimated values for concentrate were none in most cases whereas for pasture they were too low to be compatible with either the registered BWG or the voluntary intake expected for animals of the recorded BW. Availability of biomass and proportion of green leaves in it was much higher in Nov than in Aug or Sept (Table 2). The alkanes concentration was also much higher in leaves than in the rest of the components of the diet (Table 3), and this could have led to a reduced proportion of alkanes from concentrate in feces, and then to an error in the estimation of diet composition and hence intake (Dove and Mayes, 2005). Under these circumstances, the supplement could have been labelled with an external source of hydrocarbons, such as beeswax (Dove and Oliván, 1998; Dove et al., 2002). However, in a study dealing with the estimation of intake and diet composition in sheep fed mixed grain/roughage diets, Valiente et al. (2003) concluded that the n-alkane method might be used for this purpose even though the concentrations of these hydrocarbons in their experiment was low. The

Table 3 – n-Alkanes concentration (mg kg^{-1}dry matter) in green leaves, stems and dead material from *Brachiaria brizantha* cv. Marandu, and in the concentrate used in the present experiment.

	Alkanes								
	C_{23}	C_{25}	C_{26}	C_{27}	C_{29}	C_{30}	C_{31}	C_{33}	C_{35}
Concentrate	13.4	15.3	3.1	5.5	4.0	1.7	8.2	2.7	0.0
	Aug								
Green leaves	0.0	2.2	2.3	5.9	32.0	11.4	143.5	104.6	49.3
Stems	0.0	1.9	1.2	3.0	11.4	2.2	33.8	27.0	14.1
Dead material	0.0	2.6	1.8	4.6	18.9	6.1	66.4	54.9	29.5
	Sept								
Green leaves	0.0	3.4	2.5	5.7	21.0	7.3	101.5	84.6	33.7
Stems	0.0	1.5	1.9	3.8	10.9	2.4	30.4	25.0	12.8
Dead material	0.0	3.0	2.2	5.3	18.7	6.4	65.0	55.0	29.7

C_{21}, C_{24} and C_{28} were not detected; C_{32} and C_{36} have been excluded as they were dosed for purposes not related to the present experiment.

difference from the present experiment was that in their case both barley straw and grain had low contents of n-alkanes, and here leaves showed much greater values. Maybe alkanes should be incorporated into supplements when their concentration in these is scarce and much lower than that in forages, but not in other cases.

With respect to the intake and diet composition values obtained in Aug and Sept (Table 4) their accuracy in grazing conditions can only be tested by indirect methods. Energy and protein requirements of the animals used in the present experiment were calculated according to the AFRC (1993), and taking into account the BWG values shown in Table 5. The nutritive value of the *Brachiaria* was as from Rueda et al. (2003), and those of citrus pulp, cottonseed meal and urea were obtained from the AFRC (1993) tables. Estimated requirements were compatible with pasture and supplement intakes (Table 4). Animal requirements were also estimated taking into account the extra activity needed to acquire the feed in grazing conditions (SCARM, 1990). In this case, however, the increased needs were not covered by the estimated intakes (Table 4). It is possible that either these intakes were underestimated or, more likely, that the extra requirements foreseen by the SCARM (1990) did not apply in the present experiment due to the availability and quality (Table 2) of the pasture, and to the supplement intake.

There were low pasture intake estimates (1.1 kg 100 kg^{-1} BW, on average; Table 4) with no effect of supplementation frequency ($p > 0.05$). Canesin et al. (2007) also observed low pasture intakes (0.83 % BW) during the dry season in crossbred steers grazing *Brachiaria brizantha* and supplemented (1 % BW) at the same frequencies as in the present experiment.

Table 4 – Dry matter intake (DMI), and dry (DMD) and organic matter (OMD) digestibility in Nellore beef steers grazing *Brachiaria brizantha* cv. Marandu in two months and supplemented with three patterns.

	Aug			$\bar{X}$	Sept			$\bar{X}$	SEM			*p*		
	OD	SS	AD		OD	SS	AD		F	M	F*M	F	M	F*M
DMI, kg day^{-1}														
Pasture	2.9	3.3	3.3	3.2	4.6	4.0	4.1	4.2	0.185	0.145	0.503	0.948	< 0.001	0.181
Supplement	1.5	2.0	1.7	1.7	1.2	1.2	1.4	1.3	0.205	0.167	0.290	0.700	0.096	0.561
Total	4.4	4.6	5.0	4.7	5.8	5.2	5.5	5.5	0.217	0.167	0.290	0.584	0.005	0.275
DMI, kg 100 kg^{-1} BW														
Pasture	0.8	0.9	1.1	0.9	1.3	1.1	1.3	1.2	0.081	0.066	0.114	0.230	0.007	0.414
Supplement	0.4	0.6	0.6	0.5	0.4	0.3	0.5	0.4	0.079	0.061	0.105	0.487	0.181	0.698
Total	1.3	1.3	1.7	1.4	1.7	1.4	1.8	1.6	0.136	0.111	0.192	0.173	0.174	0.691
DMD, % of intake	77.0	79.7	80.3	79.0	82.7	80.7	82.3	82.0	0.010	0.008	0.015	0.584	0.035	0.291
OMD, % of intake	71.0	72.7	73.0	72.4	85.3	83.3	84.3	84.3	0.013	0.010	0.018	0.929	< 0.001	0.570

SEM: Standard error of the mean; p: Probability of the differences; OD: Supplement given once daily; SS: Supplement given once daily except Saturdays and Sundays; AD: Supplement given on alternate days. F: Effect of supplementation frequency; M: Effect of month of study; F*M: Interaction.

Table 5 – Average daily body weight gain (BWG), *Longissimus* muscle area (LMA) and backfat depth (BD) in Nellore beef steers grazing *Brachiaria brizantha* cv. Marandu in different months and supplemented with three patterns.

Frequency	Months					mean	SEM			*p*		
	July	Aug	Sept	Oct	Nov	mean	F	M	F*M	F	M	F*M
	BWG (kg day^{-1})						0.041	0.050	0.087	0.593	< 0.001	0.164
OD	0.41	0.20	0.15	0.90	0.71	0.47						
SS	0.34	0.09	0.41	0.93	0.78	0.51						
AD	0.23	0.19	0.08	0.84	0.90	0.45						
mean	0.32 b	0.16 b	0.21 b	0.89 a	0.80a							
	LMA (cm^2)						0.704	0.799	1.399	0.113	< 0.001	0.946
OD	53.3	55.1	55.7	63.3	66.7	58.8						
SS	53.0	52.8	54.3	59.1	64.5	56.8						
AD	53.5	54.9	55.0	63.9	66.2	58.7						
mean	53.2 b	54.3 b	55.0 b	62.1 a	65.8 a							
	BD (mm)						0.155	0.160	0.278	0.038	0.002	0.745
OD	3.41	3.33	3.33	4.30	4.04	3.68 A						
SS	3.23	3.18	3.15	4.48	4.79	3.76 A						
AD	2.91	3.07	3.00	3.44	3.69	3.22 B						
mean	3.18 b	3.19 b	3.16 b	4.07 a	4.17 a							

SEM: Standard error of the mean; *p*: Probability of the differences; OD: Supplement given once daily; SS: Supplement given once daily except Saturdays and Sundays; AD: Supplement given on alternate days. F: Effect of supplementation frequency; M: Effect of month of study; F*M: Interaction. a, b Different letters indicate differences ($p < 0.05$) between months; A, B Different letters indicate differences ($p < 0.05$) between supplementation frequencies.

Krehbiel et al. (1998) showed that dry matter intake and net portal and hepatic flux of nutrients in mature ewes fed low-quality forage increased in response to supplementation with 80 g CP day^{-1} in the form of soybean meal, with no differences due to the frequency of administration of the supplement (once daily or every three days). Also Huston et al. (1999) did not observe sdifferences in forage and supplement intake due to supplementation frequency, 1x week (1 % BW) or 3x week (0.45 % BW), in Hereford × Brangus adult beef cattle in winter. The opposite results were reported by Beaty et al. (1994), who observed that a reduction in supplementation frequency, from 7x (0.4 % BW) to 3x week (1 % BW), was reflected in a reduction of straw and total DM intake in Angus × Hereford steers.

Average pasture DM intake values were rather low and the question why animals did not eat more should be asked (Table 4). Time spent grazing during the day (6h00 to 18h00) was scarce (3.7 h on average, with no differences between supplementation frequencies; Table 6) and it is not likely that grazing at night would have increased this figure by more than 35 % (Dulphy et al., 1980). Probably the high proportion of stems and their low quality (Table 2) made the process of resource acquisition a negative aspect of feeding (Tolkamp and Ketelaars, 1992). Theoretically, the animals were able to eat more concentrate and hence gain more weight but Ketelaars and Tolkamp (1996) have stated that animals have their own objectives that are possibly quite different from rapid growth or high milk production. It is likely that oxygen efficiency of feeding behavior, i.e., net energy intake per liter of oxygen consumption (Ketelaars and Tolkamp, 1996) would have been less favorable if animals had increased their intake.

Estimated DM digestibility (DM and OM) was not affected (Table 4) by supplementation frequency ($p > 0.05$) but it was by the month of study (lower values in Aug than in Sept; $p < 0.05$). These results are in accordance with the higher proportions (although non-significant; Table 2) of leaves in the available pasture biomass. However, lower intakes (4.9 vs. 5.5 kg day^{-1}) and higher supplement proportions in the diet (0.35 vs. 0.24) were estimated in Aug (Table 4), and this should have resulted in higher DMD (Waldo et al., 1972). Not only were the DMD estimates odd in Aug but also those for OMD, which were abnormally lower than DMD estimates. When a closer look into the raw data was taken, it became patently obvious that estimates of OM intake in Aug included, on average, 0.15 dead material. This was the only case where intake of this fraction was estimated, as DMI estimates (in both Aug and Sept) or OM intake (OMI) estimates in Sept did not include this fraction.

Table 6 – Feeding behaviour between 06h00 and 18h00 of Nellore beef steers grazing *Brachiaria brizantha* cv. Marandu and supplemented with three different patterns.

	Frequency			SEM	p
	OD	SS	AD		
Grazing time, hours	3.93	3.92	3.33	0.157	0.098
Mean grazing time*	15h00 b	15h30 a	15h45 a	0.008	< 0.001

SEM: Standard error of the mean; OD: Supplement given once daily; SS: Supplement given once daily except Saturdays and Sundays; AD: Supplement given on alternate days; p: significance; *Time around which grazing activity was concentrated. a, b Different letters indicate differences ($p < 0.05$) between supplementation frequencies.

Dead material had the second highest level of C_{31} concentration, only after leaves (Table 3). Hence, a relatively small estimated intake of this fraction would have greatly increased the estimated concentration of C_{31} in the whole diet. That was the case in the present experiment, leading to OMD values lower than DMD in Aug. The increase in C_{31} concentration in the estimated OM intake (whole diet), with regard to concentration in DM intake, was proportionally higher than the increase in fecal concentration of C_{31} in OM with respect to concentration in DM. With regard to the lower DMD values found in Aug than in Sept, it is possible that animals actually ingested some dead material in the first month that is not revealed by the n-alkane technique.

Paraffin concentrations of diet components on a DM basis were lower than on an OM basis, probably leading to less accurate estimates of diet composition (not presented) on a DM basis than on an OM basis. Although DM intake estimates were compatible with estimated animal requirements, as discussed above, it seems that more accuracy is needed when estimating diet composition, as it has a significant effect on digestibility. Treating supplements with external alkanes should then be a common practice when the study of this variable is sought in supplemented grazing animals.

In absolute terms, digestibility values were about 2-19 % above those reported by Pereira et al. (2008) working with beef cattle fed diets containing *Brachiaria brizantha* silage and concentrate at different ratios (71 % for both DMD and OMD, and 35 % concentrate in the diet). Even though silage is expected to have lower digestibility than green forage, and the concentrate used by the above authors was composed of ingredients other than those used in the present experiment, discrepancies may reflect, one more time, the need for a more accurate use of the n-alkanes technique.

Papers dealing with the effect of supplementation frequency in animals fed tropical pastures on DM or OM digestibility are scarce. To our knowledge, this is the first attempt to estimate intake and digestibility in beef steers grazing a tropical pasture and supplemented at different frequencies using the n-alkane technique. Although this latter can, and must be improved, the results obtained were within the range considered 'normal' for the stated conditions.

Body weight gain

Supplementation frequency did not affect BWG ($p > 0.05$), although this was affected by the month of study ($p < 0.05$), with higher figures in Oct and Nov (Table 5). The higher biomass availability and proportions of green leaves in the pasture together with the higher CP and lower NDF content may explain the differences (Table 2), and also the increasing % CP of the supplement leading to increased forage intake. These results are in accordance with those reported by other authors (Huston et al., 1999; Schauer et al., 2005) who observed similar BWG in ruminants fed on low-quality forages and supplemented either infrequently or once daily.

Ruminants are efficient in maintaining adequate levels of N in the rumen, even during short periods of undernutrition, i.e. infrequent supplementation in animals fed low quality roughages (Bohnert et al., 2002). Urea recycling is known to have a relevant role. These authors stated that, even in conditions of infrequent supplementation, rumen microorganisms were able to adequately ferment structural carbohydrates then coping successfully with maintaining animal performance.

Canesin et al. (2007) recorded BWG of crossbred beef steers grazing *Brachiaria brizantha* cv. Marandu at a low stocking rate (1.32 AU ha^{-1}, on average), and supplemented once daily, once daily except Saturdays and Sundays, or on alternate days, at 1 % day^{-1} of their BW. Supplement was composed of corn, soybean meal and urea, and there were no differences in BWG between supplementation frequencies, with an average value of 0.54 kg day^{-1}.

Carcass characteristics

The area of the *Longissimus* muscle area (LMA) was similar among the supplementation frequency treatments ($p > 0.05$), and differences were observed in backfat depth (BD) ($p < 0.05$) (Table 5). BD was thinner ($p < 0.05$) in animals supplemented AD (3.22 mm), but even in this case it was within the standards considered satisfactory for a finishing steer (3-6 mm). In a study with crossbred steers grazing *Brachiaria brizantha* cv. Marandu Canesin et al. (2006) did not find differences in LMA or BD between supplementation frequencies (once daily, once daily except Saturdays and Sundays, or on alternate days), with average values slightly higher for LMA (59.7 cm^2) and lesser for BD (3.3 mm), probably due to the use of a different genetic basis (*Bos indicus* × *Bos Taurus*).

Most of the information on carcass characteristics using ultrasonography has been obtained from confined animals, and the technique has hardly been used under grazing conditions. The reasons may be found in the more aggressive behaviour and the thinner BD of free-ranging animals, which makes it difficult to obtain good images. Apart from this, Williams (2002) showed similar correlations between ultrasound and carcass measurements for backfat (r = 0.81 to 0.86) and *Longissimus* muscle area (r = 0.61 to 0.76).

Feeding behavior

Table 6 shows that grazing activity was scarce between 6h00 and 18h00, with most of the time dedicated to pasture intake occurring after noon, probably as a consequence of supplementation being offered at 08h00. Time spent grazing between 6h00 and 18h00 did not differ among supplementation frequencies ($p > 0.05$), and accounted for 3.93, 3.92 and 3.33 h for OD, SS and AD, respectively. However, the time around which grazing activity was concentrated (mean grazing time) was 15h00 for animals supplemented OD, and 15h30 and 15h45 for those on SS and AD, respectively. Differences were found only between OD and the two other supplementation frequencies ($p < 0.05$). These results outline the relationships between grazing time, and pasture intake (Table 4) and weight gain (Table 5), which were not affected by supplementation frequency either.

Schauer et al. (2005) did not find differences ($p > 0.05$) in grazing time between Angus × Hereford cows grazing native ranges in the northern Great Basin supplemented once daily or every six days (7.08 vs. 7.87 h day^{-1}), whereas the grazing time was 2.1 h longer in non-supplemented animals. These results are in agreement with those found by Beaty et al. (1994), who did not observe differences in grazing time between pregnant Angus × Hereford beef cows grazing dormant tallgrass prairie pastures supplemented daily or three times per week (6.8 vs. 7.0 h day^{-1}).

Overall, reducing the supplementation frequency to 5x weekly or on alternate days appeared to be an efficient management practice in the present experiment, as there was no reduction in alkane-estimated total intake with respect to animals supplemented daily. There were no differences either in terms of grazing time during day hours or average daily gain, even when the amount and quality of pasture were limiting. Hence, reduced supplementation frequency can be adopted with the aim of reducing labor costs.

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