Performance of sheep grazing *Panicum maximum* cv. Massai and supplemented with protein sources during the dry season

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Abstract

The low quality of tropical grasses in the dry season justifies the use of dietary supplements to meet the nutritional needs of sheep. The objective of this study was to evaluate the effect of supplementation with high-protein feed on mutton sheep performance and yield in Massai grass pastures during the dry season. The treatments corresponded to four supplements, namely *Leucaena leucocephala* leaf hay, *Gliricidia sepium* leaf hay, soybean meal, and a treatment that received only multiple mixture (protein salt) ad libitum. The pasture was evaluated for canopy height, forage supply, and chemical composition of leaf blade, stem and dead material. Average daily gain, weight gain per area, and the stocking rate were also evaluated. There were four grazing cycles because the grazing method was rotational stocking with seven days of occupation and 35 days of rest. The interaction between supplements and grazing cycles was not significant, and no effects of the supplements were found for any of the variables. The highest forage and leaf blade offerings were observed in the first grazing cycle. The highest levels of crude protein and lower neutral detergent fibre and lignin of pasture components were observed in cycles 3 and 4. The highest average daily gains per animal and per area were observed in cycles 1 and 2. Protein supplementation of sheep in Massai grass pastures promotes satisfactory gains during the dry season, and *Leucaena* and *Gliricidia* hays can be used as protein sources instead of soybean meal.

Keywords: *Gliricidia sepium*, leguminous, *Leucaena leucocephala*, pasture

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Introduction

Tropical regions are characterized by great variation in the quantity and quality of pastures throughout the year, with an abundance of forage in the rainy season and a shortage in the dry season. This causes seasonality in animal production, which can lead to economic losses for the rural producer, since the animals take longer to reach slaughter weight and causes fluctuations in the supply of animal products (Bayão et al., 2016).

Animal production systems that use forage species which are adapted to the climatic conditions of the region and are resistant to periods of drought periods can help to improve efficiency of production (Felix et al., 2016). *Panicum maximum* cv. Massai has been shown to be resistant and adapted to regions with low precipitation because of its rapid establishment, and tolerance of infertile soils and infestation by leafhoppers (Rodrigues et al., 2014).

During the dry season, the supply of potentially digestible fibre in the forage is satisfactory, with the protein deficit being mostly responsible compromising the performance of grazing animals. Use of appropriate supplements may increase production rates (Reis et al., 2009). Costs of supplementation can impinge on this management strategy, and therefore their use must be strategic. Finding alternative options in the region could reduce production costs.

Evaluating species that are adapted to the edaphoclimatic conditions of the region for their suitability as dietary supplements is important to capture economic benefits from grazing animals in tropical regions.
Leucaena (Leucaena leucocephala) and Gliricidia (Gliricidia sepium) are leguminous plants with high potential for hay production. These hays can be used as sources of protein in diets of ruminant animals, since they contain 18% to 30% crude protein (CP), as well as being adapted to cultivation in soils of medium to low fertility (Edwards et al., 2012). Thus, the objective of this study was to evaluate leguminous hays and soybean meal as sources of supplemental protein on performance of 3-month old lambs grazing Massai grass pastures during the dry season.

Materials and Methods
The experiment was conducted at the experimental area of the Forage Study Group which is located in the Academic Unit Specialized in Agricultural Sciences of the Agricultural School of Jundiaí, Macaíba Campus, Federal University of Rio Grande do Norte, Macaíba RN, Brazil. The coordinates of the experimental area are 5° 53' 34'' latitude south, 35° 21' 50'' longitude west, and the altitude is 50 metres. According to Thornthwaite’s (1948) classification, the climate is sub-humid dry with excess water from May to August. The historical average annual rainfall is 1048 mm, with an average annual accumulated potential evapotranspiration of 1472 mm.

The experiment was conducted between November 2012 and April 2013. Temperature data were obtained from the database of National Institute of Meteorology, and a stainless steel Ville de Paris pluviometer was installed on the experimental site to obtain precipitation data (Figure 1).

Figure 1 Precipitation and temperatures from January 2012 to December 2013 at the experimental area of the Forage Study Group, Federal University of Rio Grande do Norte

Chemical characteristics of the soil (Quartzipsamment) at the experimental area of the Forage Study Group, Federal University of Rio Grande do Norte were obtained from laboratory analyses. Based on the results (Table 1), 50 kg/ha phosphorus pentoxide was applied to increase the phosphorus content of the soil, according to the recommendation by Ribeiro et al. (1999).

Table 1 Chemical characteristics of the soil at the experimental area of the Forage Study Group, Federal University of Rio Grande do Norte at depths of 0-20 cm and 20-40 cm

| Layers (cm) | P | K | Na | pH | Ca | Mg | Al | H+Al | BS (%) |
|------------|---|---|----|----|----|----|----|------|-------|
| 0–20       | 7.0| 75.0| 27.0| 6.1| 1.2| 0.5| 0.0| 1.6   | 55.7  |
| 20–40      | 4.0| 124.0| 29.0| 5.3| 1.6| 0.8| 0.1| 2.3   | 55.3  |

P: phosphorus, K: potassium, Na: sodium, Ca: calcium, Mg: magnesium, Al: aluminium, H: hydrogen, BS: base saturation

The Massai grass was planted in the first half of 2011 with viable seed sown at a rate of 4 kg per hectare using a conventional row seeding system. The grassland was fertilized with 50 kg/ha N in the form of ammonium sulfate 30 days before the start of the experiment to increase the forage accumulation during the dry season and to allow for similar forage production in the paddocks.

The experimental area was divided into four blocks of 0.24 ha. Each block was subdivided into six paddocks of 0.04 ha. The blocks were occupied by sheep and they were supplemented with multiple mixture...
(MM), MM and Leucaena leaf hay (MM+LH), MM and Gliricidia leaf hay (MM+GH), or MM and soybean meal (MM+SM) (Table 2). The dietary supplements were isonitrogenous. The supplement was offered at a level that met 25% CP needs in sheep, according to the National Research Council (2007). The MM was composed of 55% mineral supplement, 25% ground corn and 20% farming urea. The paddocks were stocked intermittently with fixed periods of occupation and rests of seven days and 35 days, respectively. Four grazing cycles were evaluated with a total experimental period of 168 days.

**Table 2** Chemical composition of supplements for sheep grazing in Massai grass pastures (g kg DM⁻¹)

| Supplement         | DM  | Ashes | CP  | NDF | ADF | Lignin |
|--------------------|-----|-------|-----|-----|-----|--------|
| Leucaena hay       | 894 | 95    | 210 | 490 | 301 | 167    |
| Gliricidia hay     | 833 | 109   | 180 | 398 | 288 | 122    |
| Soybean meal       | 896 | 67    | 460 | 159 | 97  | 16     |
| Multiple mixture   | 970 | 554   | 585 | 32  | 18  | 0.3    |

DM: dry matter; CP: crude protein; NDF: neutral detergent fibre; ADF: acid detergent fibre

Twenty-four Santa Ines male sheep (six per module) with an average initial weight of 15.1 ± 2.8 kg (MM), 15.0 ± 2.2 kg (LH), 15.5 ± 2.8 kg (GH), and 14.8 ± 2.5 kg (SM), and average initial age of three months were used as defoliating agents. The sheep were kept in pasture from 07h30 to 16h30 and then put into a covered sheepfold where they were supplemented and kept overnight. There were four stalls in the sheepfold for the four groups, each measuring 9 m², and they were provided with feeder, water dispenser and salt.

Animals were weighed at the beginning of the experiment and at the end of each grazing cycle to obtain average daily weight gain (g/day). Stocking rate per grazing cycle (animal units per hectare (AU ha⁻¹)) was calculated as the product of average animal weight and number of days they remained in the module, according to Petersen & Lucas Jr. (1968), where an AU corresponds to 25 kg of live weight (LW). Total live weight gain per hectare (kg LW ha⁻¹) was obtained by calculating the product of daily weight gain and the stocking rate in each grazing cycle.

Forage mass (FM) in the pre-grazing condition was obtained in four representative areas in four of the six paddocks of each module by cutting the forage close to the ground using a square of 0.5 m². The forage allowance (FA) was calculated as the product of average animal weight and number of days they remained in the module, according to Petersen & Lucas Jr. (1968), where an AU corresponds to 25 kg of live weight (LW). Total live weight gain per hectare (kg LW ha⁻¹) was obtained by calculating the product of daily weight gain and the stocking rate in each grazing cycle.

Forage mass (FM) in the pre-grazing condition was obtained in four representative areas in four of the six paddocks of each module. The morphological components of the pasture in the pre-grazing period were obtained from two combined samples, which were separated manually into leaf blade, stem (stem + sheath) and dead material. The morphological constituent samples were ground to 1 mm and analysed for CP content, in vitro organic matter digestibility (IVOMD) percentages, neutral detergent fibre (NDF) and lignin contents using the near-infrared reflectance spectrophotometer (NIRS) system, according to the procedures described by Marten et al. (1985). For NIRS analyses, the curves were calibrated on a Foss NIR Systems model 5000 (FOSS, Hilleroed, Denmark) using ISI WINISI II Project Manager Software V1.02.

Forage disappearance was calculated by the difference between pre- and post-grazing forage mass. Canopy height was obtained in pre-grazing and post-grazing periods using a graduated ruler in centimetres at 40 representative points.

At the start of the experiment, both the animals and paddocks were relatively uniform. Thus, the experimental design was completely randomized with four treatments (supplements) and six replications (paddocks or animals). Treatments were arranged in a split plot in which the supplements were allocated in the plot and grazing cycles in the subplots. Data were submitted to analysis of variance, and means were compared with Tukey test with a 5% level of significance using Sisvar, version 5.6 (Ferreira, 2011). The linear model used to analyse the data was:

\[ Y_{ijk} = \mu + s_i + a_{ij} + c_k + (cs)_{ik} + e_{ijk} \]
where: \( Y_{ijk} \) = an observed value for supplement \( i \), cycle \( k \) and repetition \( j \); \( \mu \) = the overall mean; \( s_i \) = the effect of supplement \( i \) (\( i = \text{MM, LH, GH, or SM} \)); \( \alpha_{ij} \) = random whole-plot error; \( c_k \) = effect of grazing cycle \( k \), (\( k = 1-4 \)); \( (cs)_{ik} \) = the interaction effect of supplement \( i \) and cycle \( k \); and \( \varepsilon_{ijk} \) = random sub-plot error.

**Results**

Supplements that were provided to the lambs did not affect any of the variables in the pasture (\( P >0.05 \)) (Table 3), nor was the interaction of supplements with grazing cycles significant. Significant differences were observed only between grazing cycles. Greater forage and leaf blade allowances were observed in the first grazing cycle (\( P <0.05 \)), while the forage allowance decreased by 41.1\% from the first to the fourth grazing cycle.

**Table 3** Average forage allowance (kg DM/100 kg LW), forage disappearance (kg/ha DM) and pre- and post-grazing canopy height (cm) in Massai grass pastures grazed by mutton sheep during the dry season in function of protein supplements

| Variable                        | Supplements | SEM  |
|---------------------------------|-------------|------|
|                                 | MM          | LH   | GH   | SM  |
| Forage allowance                | 116.3       | 100.7| 104.7| 95.7| 4.3 |
| Leaf blade                      | 25.3        | 27.5 | 30.7 | 27.6| 2.5 |
| Stem                            | 12.1        | 9.0  | 12.6 | 11.2| 1.0 |
| Dead material                   | 78.5        | 65.6 | 61.2 | 57.8| 1.0 |
| Forage disappearance            | 2347.1      | 2017.1| 2222.5| 2262.5| 234.0|
| Green forage disappearance      | 1100.4      | 1159.2| 1382.8| 1252.6| 142.7|
| Canopy height (pre-grazing)     | 27.4        | 22.8 | 22.6 | 22.5| 0.9 |
| Canopy height (post-grazing)    | 20.4        | 17.6 | 19.2 | 17.6| 1.2 |

\( a, b, c, d \) Row means with different superscripts differ significantly at \( P <0.05 \)

MM: multiple mixture; LH: Leucaena hay; GH: Gliricidia hay; SM: soybean meal

Stem allowance was greater (\( P <0.05 \)) in the first two cycles and less in the last two, while the allowance of dead material was reduced (\( P <0.05 \)) only in the fourth cycle compared with cycles 1 and 2. Total forage disappearance did not differ between the grazing cycles (\( P >0.05 \)) with an average value of 2166.5 kg/ha DM, while green forage disappearance was reduced in the third grazing cycle. Pre- and post-grazing pasture heights decreased with the advance of grazing cycles (Table 4).

The grazing cycle affected the chemical composition of leaf blades, stem and dead material, except for in vitro organic matter digestibility (IVOMD) and lignin of dead material which had overall means of 359 and 83 g/kg DM, respectively (Table 5). Neutral detergent fibre and lignin concentrations of the stem were reduced in the fourth grazing cycle (672 g/kg DM and 42 g/kg DM, respectively) compared to the previous cycles. The concentration of CP in dead material increased during the fourth grazing cycle. An inverse trend in NDF levels of the dead material was observed with mean values of 763 g/kg DM in the first three cycles compared with 727 g/kg DM in the fourth cycle (Table 5).
Table 4 Average forage allowance and components (kg DM/100 kg LW), total forage and green forage disappearance (kg/ha DM) and pre- and post-grazing canopy height (cm) in Massai grass pastures grazed by mutton sheep supplemented during the dry season

| Variable                                      | Grazing cycles | SEM |
|-----------------------------------------------|----------------|-----|
|                                               | 1  | 2  | 3  | 4  |     |
| Forage allowance                              | 138.5<sup>a</sup> | 111.2<sup>b</sup> | 86.3<sup>c</sup> | 81.6<sup>c</sup> | 4.3 |
| Leaf blade                                    | 50.3<sup>a</sup> | 24.0<sup>b</sup> | 15.1<sup>b</sup> | 21.9<sup>b</sup> | 2.5 |
| Stem                                          | 16.2<sup>a</sup> | 13.7<sup>a</sup> | 7.6<sup>b</sup>  | 7.3<sup>b</sup>  | 1.0 |
| Dead material                                 | 71.8<sup>a</sup> | 74.2<sup>a</sup> | 65.1<sup>ab</sup> | 52.1<sup>b</sup> | 4.3 |
| Forage disappearance                          | 2289.8<sup>a</sup> | 2548.4<sup>a</sup> | 1846.3<sup>a</sup> | 1981.5<sup>a</sup> | 234.0 |
| Green forage disappearance                    | 1541.6<sup>a</sup> | 1340.2<sup>a</sup> | 808.3<sup>b</sup> | 1328.9<sup>a</sup> | 102.7 |
| Canopy height (pre-grazing)                   | 30.8<sup>a</sup> | 25.7<sup>b</sup> | 21.1<sup>c</sup> | 17.7<sup>d</sup> | 0.9 |
| Canopy height (post-grazing)                  | 26.7<sup>a</sup> | 18.9<sup>b</sup> | 14.6<sup>c</sup> | 13.2<sup>c</sup> | 0.1 |

<sup>a,b,c</sup> Row means with different superscripts differ significantly at *P* <0.05

Table 5 Chemical composition and digestibility of the morphological components of Massai grass pastures at pre-grazing

| Variable (g/kg DM)                           | Grazing cycle | SEM |
|----------------------------------------------|---------------|-----|
|                                              | 1  | 2  | 3  | 4  |     |
| Leaf blade                                   |               |     |     |     |     |
| Crude protein                                | 91<sup>b</sup> | 82<sup>b</sup> | 133<sup>a</sup> | 122<sup>a</sup> | 3.9 |
| Neutral detergent fibre                      | 721<sup>a</sup> | 726<sup>a</sup> | 702<sup>b</sup> | 660<sup>c</sup> | 5.1 |
| In vitro organic matter digestibility        | 612<sup>b</sup> | 581<sup>b</sup> | 700<sup>a</sup> | 687<sup>a</sup> | 15.7 |
| Lignin                                       | 55<sup>a</sup> | 57<sup>a</sup> | 38<sup>a</sup> | 38<sup>a</sup> | 2.1 |
| Stem                                         |               |     |     |     |     |
| Crude protein                                | 60<sup>abc</sup> | 53<sup>c</sup> | 74<sup>a</sup> | 68<sup>ab</sup> | 3.8 |
| Neutral detergent fibre                      | 711<sup>a</sup> | 705<sup>a</sup> | 710<sup>a</sup> | 672<sup>b</sup> | 7.1 |
| In vitro organic matter digestibility        | 586<sup>ab</sup> | 572<sup>b</sup> | 595<sup>ab</sup> | 608<sup>a</sup> | 11.4 |
| Lignin                                       | 59<sup>a</sup> | 58<sup>a</sup> | 54<sup>a</sup> | 42<sup>b</sup> | 2.0 |
| Dead material                                |               |     |     |     |     |
| Crude protein                                | 45<sup>b</sup> | 42<sup>b</sup> | 44<sup>b</sup> | 55<sup>a</sup> | 2.0 |
| Neutral detergent fibre                      | 765<sup>a</sup> | 754<sup>a</sup> | 762<sup>a</sup> | 727<sup>b</sup> | 2.0 |
| In vitro organic matter digestibility        | 350<sup>a</sup> | 357<sup>a</sup> | 361<sup>b</sup> | 370<sup>a</sup> | 5.6 |
| Lignin                                       | 86<sup>a</sup> | 79<sup>a</sup> | 83<sup>a</sup> | 85<sup>a</sup> | 1.5 |

<sup>a,b,c</sup> Row means with different superscripts differ significantly at *P* <0.05
No effects of the dietary supplements ($P > 0.05$) were observed for average daily gain per animal, weight gain per unit area or stocking rate (Table 6).

**Table 6** Performance of sheep supplemented with protein sources in Massai grass pastures by supplement type

| Variable                          | Supplements | SEM |
|----------------------------------|-------------|-----|
|                                  | MM          | LH  | GH  | SM  |     |
| Average daily gain (g/day)       | 69.2        | 71.6| 65.3| 72.7| 3.2 |
| Stocking rate (AU/ha)            | 20.6        | 20.3| 20.1| 21.8| 0.3 |
| Weight gain per area (g/ha/day)  | 1425.5      | 1453.5| 1312.5| 1584.9| 134.6|

MM: multiple mixture; LH: Leucaena hay; GH: Gliricidia hay; SM: soybean meal; AU: animal unit of 25 kg

In comparing grazing cycles, the highest weight gain per animal and per unit area ($P < 0.05$) were observed in cycles 1 and 2 (Table 7), while the stocking rate increased by 38% from the first to the fourth cycle ($P < 0.05$).

**Table 7** Performance of sheep supplemented with protein sources in Massai grass pastures by grazing cycle

| Variable                          | Grazing cycle | SEM |
|----------------------------------|---------------|-----|
|                                  | 1  | 2  | 3  | 4  |     |
| Average daily gain (g/day)       | 94.6$^a$     | 91.6$^a$| 56.3$^b$| 59.7$^b$| 3.2 |
| Stocking rate (AU/ha)            | 17.7$^c$     | 19.8$^b$| 20.8$^b$| 24.5$^a$| 0.3 |
| Weight gain per area (g/ha/day)  | 1674.9$^a$   | 1813.7$^a$| 1169.0$^b$| 1462.3$^b$| 134.6|

$^{a,b,c}$ Row means with different superscripts differ significantly at $P < 0.05$

AU: animal unit of 25 kg

**Discussion**

Similarities in the forage mass and structure of the pastures as a function of the supplements show uniformity in the treatment application (Table 3). The observed average forage allowance values were greater than the 36.9 kg DM/100 kg LW observed by Garcia et al. (2014) for cattle supplemented in the dry season. These authors emphasized the lack of any decrease in pasture consumption by the animals at this forage allowance. According to Fernandes et al. (2017), this forage mass and structure is satisfactory for production of grazing animals during the dry period.

Reductions in forage yields, morphological components and canopy heights over the passage of each grazing cycle (Table 4) can be attributed to the very limited pasture growth. This lack of forage production was expected as the experiment was carried out in the dry period. Along with the water deficit, pasture regrowth was not sufficient to restore the forage mass consumed by the animals during the periods when the paddocks were grazed. Emerenciano Neto et al. (2017) recommended that Massai grass to be grazed by sheep have a canopy between 35 and 40 cm in height prior to grazing. In this study, these values were approached only in the first cycle (30.8 cm).

Leaf blade allowances in cycles 2 and 4 were similar to the 27 kg DM/100 kg LW that was observed Euclides et al. (2008) for Massai grass pastures in the dry period. However, in cycle 1 of the present study, the leaf blade allowance less than in the study of Euclides et al. (2008) due to differences in stocking between the studies. In this study stocking was fixed, whereas in Euclides et al. (2008) the number of animals and consequently the forage allowance was adjusted to the available forage mass.

The similarity of forage disappearance across the grazing cycles may be indicative of a lack of variation in total forage consumption by the animals. The reduced green forage disappearance in cycle 3 of may be a product of the unavailability of this component resulting of preferential consumption in the previous cycles. In cycle 4 contemporaneous rains (Figure 1) allowed renewed forage growth.
The highest CP and the lowest NDF and lignin levels of pasture components were observed in cycles 3 and 4. These results can be explained by modification of structure of the pastures due to the precipitation (Figure 1) that occurred late in the experimental period and the consequent increase in regrowth of the Massai grass with its improved nutritional value.

In vitro organic matter digestibility and CP of the leaf blade in cycles 1 and 2 were similar to the 523 g/kg DM and 80 g/kg DM as found by Euclides et al. (2008) for the same cultivar in the dry period. These authors observed reduced animal performance when grazing Massai grass when compared with the grazing of Mombasa grass. This reduction in performance resulted from the lower IVOMD and CP of Massai grass. This highlights the importance of using protein supplements for animals kept in Massai grass pastures. Further, the anatomical structure of Massai grass with its increased presence of Girder I structure compared to Mombasa grass (Lempp et al., 2000). According to Paciullo (2002), this structure hinders epidermis detachment from the rest of the leaf, which promotes greater resistance to mechanical and chemical damage in the digestion process.

The lack of supplemental effect on animal performance (Table 6) can be attributed to the equal amount of protein provided by each supplement, although the nature of the protein source is different. This result is positive from an economic point of view, since producing and using such hays can promote increases in production efficiencies. The average daily gains achieved using these supplements may facilitate the avoidance of seasonality in animal production because the growth rates observed here were greater than the 49.2 g/day and 41.6 g/day observed by Emerenciano Neto et al. (2014; 2018) for unsupplemented sheep grazing Massai grass pastures during the rainy season and under irrigation, respectively.

The reduced weight gain per animal and per area observed after the second grazing cycle (Table 7) may be related to lower forage allowance (Table 5). According to Silva et al. (2016), animals in pastures with low forage allowance may suffer food restriction and therefore increase their grazing time, requiring more time searching for the more nutritious portions of the plant. Forage consumption or animal performance is increased based on increased forage allowance (Hodgson, 1990). Moreover, protein supplementation is effective only when dry matter is available, and energy supplementation is necessary if this is not the case (Silva et al., 2008).

The increase in the stocking rate was a result of the increase in the live weight of the animals because fixed stocking was used. The results obtained in this study were satisfactory, as Emerenciano Neto et al. (2014) observed rates of 26.6 AU/ha in Massai grass pastures in the rainy season. These reported values are close to those Growth rates achieved in in cycle 4 of this study, which occurred during the dry season, were similar to those of Emerenciano Neto et al. (2014) wherein only grass was used as a food source for the animals.

Conclusion

Protein supplementation of lambs in Massai grass pastures promotes satisfactory individual and per unit area production during the dry season, provided that forage supply is adequate. Leucaena and Gliricidia hays can be used instead of soybean meal as a source of dietary protein for growing sheep kept in Massai grass pastures. This is justified by the similarity of growth rates by animals supplemented with these hays to those that were supplemented with soybean meal. The similarity of animal production per unit area, regardless of the protein source, also supports this conclusion.

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Authors' Contributions

Conception and design: LSF, GSD and JVEN; data collection and analysis: LSF, IMMA, ELLV and JVEN; drafting of paper: LSF; critical revision and final approval of version to be published: GSD, JVEN and MGC.

Conflict of Interest Declaration

The authors certify that they have no affiliations with any organization or entity with financial or non-financial interest in the subject matter or materials discussed in this manuscript.

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