

Original Research Article
**MORPHOLOGICAL AND PRODUCTIVE EVALUATION OF
MOMBASA GRASS AS A FUNCTION OF PHOSPHATE,
NITROGEN, AND BIOSTIMULANT APPLICATION**

Abstract

Nitrogen when applied to the surface suffers losses by volatilization. Therefore, one of the ways to reduce this loss is to use substances that delay the hydrolysis of NH_3 . The objective of this study was to evaluate the physiological characteristics of Mombasa grass as a function of nitrogen sources associated or not with volatilization inhibitors (ASP4) and nitrification (CTN). The experiment was conducted in a greenhouse, UFT - Gurupi, in DBC, with six treatments and five replications, being: SN1 - Sulfammo without ASP4; SN3 - Sulfammo + 6kg/ton of ASP4; NC1 - Sulfammo + CTN without ASP4; NC3 - Sulfammo + CTN + 6kg/ton of ASP4, URE - Urea and control without nitrogen. It is note point that all treatments received 100 kg ha^{-1} of N. The following parameters were evaluated: Chlorophyll A, Chlorophyll B, Total Chlorophyll, internal CO_2 concentration, transpiration rate, stomatic conductance, CO_2 assimilation rates, water use efficiency, and instantaneous efficiency of ribulose enzyme (RUBI) carboxylation Data were analyzed by MANOVA, using the main component technique (PCA) using the R[®] 3.5 software. According to the PCA scores, the productive characteristics are CloB; CloA; CloTO. ASSI, RUBI showed the highest variations, all positive, both in PC1 and PC2. The treatments that most influenced the characteristics were NC1 and NC3, demonstrating an inverse tendency to the controls. Crop development was significantly influenced by urea and sources with and without inhibitors.

Keywords: Multivariate analysis; Sulfammo MeTA; Forage Grass.

Introduction

Brazil is the second largest cattle producer in the world, with an average herd of 232,540 million head. Most of the herd is grazed and occupies an average area of 173 million hectares. Largely created under the pasture of the genus *Urochloa* [1, 2]. However the grass *Megathyrsus maximus* cv. Mombasa has been introduced in place of these pastures, mainly due to the high productive potential, and high nutritional and protein value (12 and 16%) [3, 4].

However, part of these pastures has been suffering a production decline due to the decrease in soil fertility due to nutrient extraction and non-replacement [4]. As a matter of time, investment in fertilizers must be considered in all cases [5]. Nitrogen is

one of the main nutrients related to the maintenance of forage grass productivity, is a constituent of proteins, and is directly linked to photosynthetic processes [6, 7].

Urea (RH) is the most widely used nitrogen (N) source in world agriculture [8]. However, when applied on the surface, it suffers N losses due to ammonia volatilization (NH₃) [9]. Therefore, the best way to reduce losses is to use substances that instill the activity of urease to delay the hydrolysis of urea and reduce the loss of N [10, 11].

Material and Methods

The study was conducted in an experimental area of the Federal University of Tocantins - Gurupi in DBC, with five replicates and six treatments, being: SN1 - Sulfammo (0kg/ton ASP4); SN3 - Sulfammo (6kg/ton ASP4); NC1 - Sulfammo + CTN (0kg/ton ASP4); NC3 - Sulfammo + CTN (6kg/ton ASP4), URE - Pure Urea and control without N application.

Yellow, Red Latosol were used, to determine the available nutrient contents, and percentages of sand, silt, and clay in the Soil Laboratory – UFT - Gurupi according to Embrapa [12] methodology. Limestone, gypsum, and basic planting fertilization were applied according to the Soil Fertility Commission of the State of Minas Gerais [13].

Table 1. Chemical and textural analysis of the Red-Yellow Latosol. Gurupi-TO, 2021.

Ca	M	Al	H+A	SB	CT	K	P	M.O	pH	San	Silt	Cla	V
-----cmol _c dm ⁻³ -----						mg dm ⁻³		%	CaCl	-----g kg ⁻¹ -----			
0,	0,4	0,	2,5	1,0	3,55	1	0,	1,3	4,9	475	50	475	30

In addition, there were three cover fertilizations with K₂O and N (Sulfammo and urea) in coverage for each uniformity cut. The forage used was *Megathyrsus maximus* cv. Mombasa.

At 30, 60, 90 days after cutting, evaluations of the morphological characteristics of forage were performed: plant height (AP, cm) - measuring with a ruler graduated in cm, the length between the soil surface to the highest end of the leaves; the number of erm (NP); leaf area (AF, cm²) will consist of the removal of 10 leaf discs with a leaker with an area of 0.38 cm². The fresh leaves were weighed on an analytical scale. The leaf area was calculated by the formula (AF = PF x AD/PD, where: PA is the leaf area estimated by the method; PF is the fresh mass of the leaf; AD is the known area of the disk, and PD is the fresh mass of the discs) according to studies conducted by Huerta; Alvim (1962) and Gomide et al. (1977) [14, 15]. The dry mass of forages (MSPA, g) was obtained from three cuts made at 30 cm above the soil level for Mombasa soon

after evaluation of the productive, physiological and morphogenic characteristics. The aerial part of the plants was collected and packed in paper bags, sent to the laboratory, and dried in a forced air circulation greenhouse, at 55° C for 72 hours.

To evaluate morphogenesis, a tiller was marked shortly after each cut, measured every 7 days. With the aid of a ruler was measured the length of the stem, and live leaves were. From these data, the calculations of the following characteristics were performed: Leaf appearance rate (TApF, leaf/child/day); Phyllochron in days (PHYLUm, days/leaf/tiller); Leaf elongation rate (TALF, cm/tiller /day); Pseudostem elongation rate (TAPC, cm/tiller/day); Lifetime of leaves (VFD, days).

The photosynthetic activity was analyzed using the IRGA - Infra Red Gas Analyser equipment, Li-Cor model LI-6400. The rates of CO₂ assimilation (ASSI), transpiration rate (TRANS), stomatic conductance (COND), internal CO₂ concentration (COIN), water use efficiency (US), instantaneous efficiency of ribulose enzyme (RUBI) were evaluated. The relative chlorophyll content was determined by the Portable SPAD-502 meter.

The data were submitted for multivariate analysis, using the technique of main components [16]. Statistical analysis and graphs were plotted using software R version[®] 3.5 [17].

Results and Discussion

In the study, the occurrence of alterations in the main growth parameters of mombasa grass was evidenced, among which it is possible to highlight the best development in mass production as a function of the application of phosphate fertilization, nitrogen, and biostimulants.

The extraction of soil nutrients by forage occurs in larger quantities than those demanded by grain production crops, in which the crop remains to remain in the cultivation area [5]. The lack of nutritional replacement increases the levels of degradation in pasture areas, even in species that adopted medium and high technological levels, such as Mombasa, so the importance of fertilization at appropriate times is important.

To choose the number of components to be evaluated, the Johnson and Wichern (1998) [18] methodology was used, where a minimum cumulative percentage close to 80% is required to explain the total variance of the data and determine the appropriate number of principal components. Thus, by accepting four components it is possible to

explain 78.10% of the data (Table 2).

Table 2. Main components (CPs), autovalues (λ_i), percentage of variance and cumulative proportion (%).

Main Component	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8	PC9	PC10
Autovalues	10.34	5.01	3.38	2.34	1.44	1.02	0.71	0.63	0.52	0.50
Percentage (%)	38.31	18.57	12.52	8.68	5.35	3.78	2.64	2.34	1.96	1.85
Cumulative percentage (%)	38.31	56.88	69.41	78.10	83.45	87.24	89.89	92.23	94.19	96.05

The first component explains 38.31% of the total variation (Table 2) and can be considered the holder of the highest correlation coefficients between the following variables: MSPA (0.95); AF (0.94); TAF (0.95) and TALF (0.81) with positive correlations with each other (Table 3).

The second component corresponds to 18.57% of the data (Table 2), where the most stand out correlation coefficients are: ALT (0.29); MPER (0.29); CloB (0.67); CloA (0.48); CloTO (0.57); TRANS (0.79); COND (0.69) and ASSI (0.49), all positive to each other (Table 3).

The third component corresponds to 12.57% of the data (Table 2), where the correlation coefficients that stand out the most are: ALT (0.30); CloB (0.34); CloA (0.27); CloTO (0.31); USA (0.26); ASSI (0.27); RUBI (0.81) and EIUA (0.66) (Table 3).

The fourth component corresponds to 8.68% of the data (Table 2), where the most stand out correlation coefficients are: RAF (0.55); TAPF (0.39); DFV (0.30); NFP (0.21); COND (0.41); SO (0.55); (0.26) and RUBI (0.35) (Table 3).

Table 3. Correlation coefficients of variables with first five components

Vari	ALT	PERF	MSPA	AF	RAF	MPER	TAF	TAL	REND	TAPF	FILO	TALF	TAPC	DFV
PC 1	0.61	0.68	0.95	0.94	-0.34	0.78	0.95	0.79	0.79	0.73	0.58	0.81	0.49	0.66
PC 2	0.29	-0.35	-0.03	-0.02	-0.06	0.29	-0.03	-0.17	-0.15	-0.42	0.00	-0.27	-0.04	-0.25
PC 3	0.30	0.06	0.01	-0.01	-0.12	0.04	0.01	-0.18	0.06	-0.23	0.04	-0.17	-0.49	-0.10
PC 4	-0.38	0.09	-0.29	-0.14	0.55	-0.41	-0.29	-0.02	-0.54	0.39	0.29	0.19	-0.06	0.30
PC 5	-0.06	-0.26	0.02	0.11	0.35	0.17	0.02	0.23	-0.09	0.08	-0.68	0.27	-0.07	-0.56
Vari	NFP	CFLF	CFPC	CloB	CloA	CloTO	COIN	TRANS	COND	ASSI	EUA	RUBI	EIUA	--
PC 1	0.58	0.79	0.12	0.44	0.70	0.65	0.01	0.14	0.43	0.49	0.21	0.30	-0.08	--
PC 2	-0.64	-0.37	-0.64	0.67	0.48	0.57	0.54	0.79	0.69	0.49	-0.54	-0.08	-0.68	--
PC 3	-0.34	-0.25	-0.50	0.34	0.27	0.31	-0.77	-0.26	-0.23	0.27	0.54	0.81	0.66	--

PC 4	0.21	0.11	0.17	0.30	-0.03	0.08	-0.03	0.14	0.41	0.55	0.26	0.35	-0.08	--
PC 5	0.16	0.24	0.17	0.25	0.02	0.10	-0.14	0.16	-0.12	0.01	-0.17	0.12	0.13	--

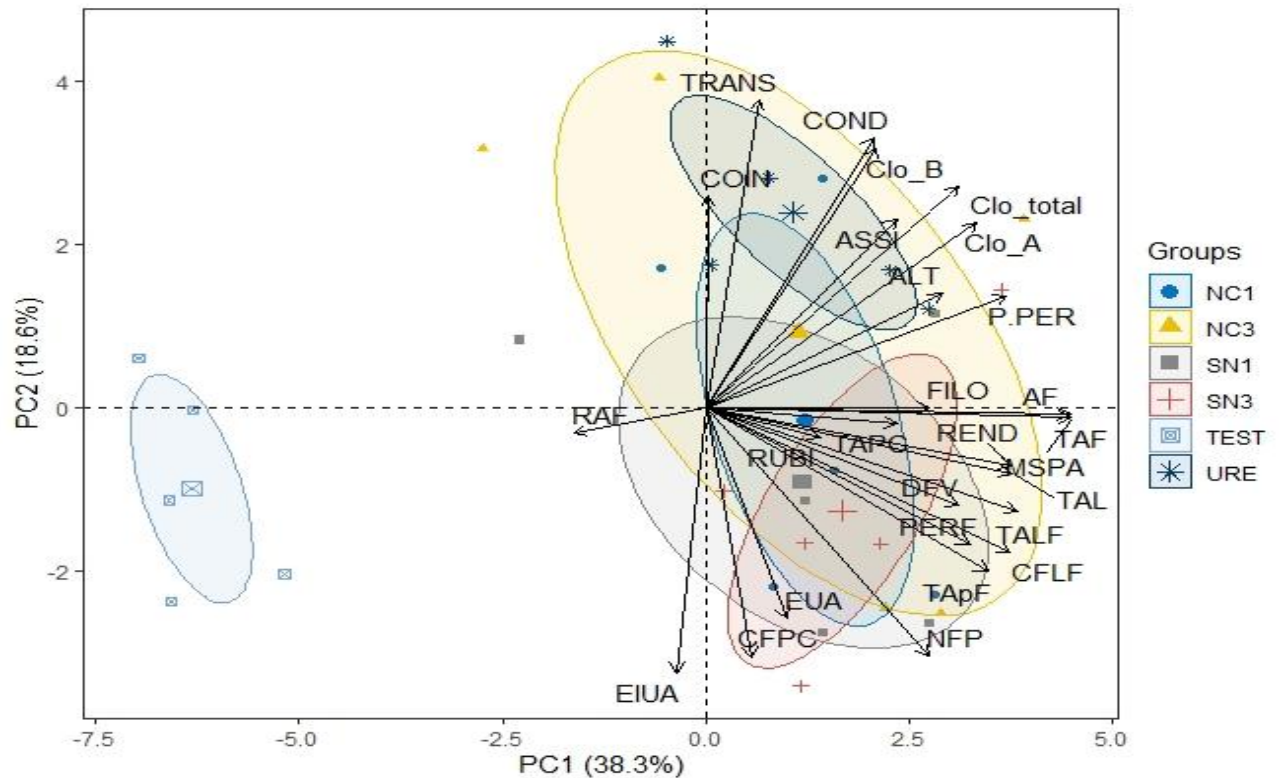
The treatments that provided the greatest influence on PC1, according to their scores and in increasing order were: SN3 (2.2078) with 6 kg/ton of ASP4; URE (1.8398), and SN1 (1.7813) without ASP4. The control behaved opposite to treatments with urea and Sulfammo with and without urease inhibitor with (-8.2259) (Table 4). The rate of hydrolysis of urea by the urease enzyme is more expressive during the first days after fertilization [19, 20]. Some studies have reported N-NH₃ losses of up to 70% of the N applied, with an average between 20 and 30%, under experimental conditions [21, 22].

Table 4. Scores of treatments under the five main components.

Treatments	PC 1	PC 2	PC 3	PC 4	PC 5
SN1	1.7813	1.7490	1.7132	-1.2282	0.8055
SN3	2.2078	2.3416	1.0724	1.2163	-0.8240
NC1	1.0985	0.9033	-2.7341	1.0817	0.5584
NC3	1.2985	-0.9218	-1.4132	-2.0037	-0.6356
URE	1.8398	-4.1305	1.0552	0.8543	0.1373
TEST	-8.2259	-0.0585	0.3066	0.0797	-0.0418

In the second component, the featured treatments and scores in ascending order were: SN3 with 6 kg/ton of ASP4 (2.3416) and SN1 (1.7490) without ASP4. The ERU and TEST behaved negatively with their scores (-4.1305) and (-0.0585). Some cultural practices help to minimize losses due to volatilization of N, such as the use of urease inhibitors [23]. Because they can reduce urea hydrolysis by inhibiting urease activity resulting in lower volatilization losses [24, 25].

Figure 1: Biplot CP1 x CP2 on the variable responses of Mombasa grass using nitrogen sources associated or not with urease inhibitors.



The negative control presented a low TEST score (-8.2259) (Table 3). The presence of nitrogen increased total chlorophyll in the plant, with this CO₂ assimilation, transpiration, and stomatic conductance also grew to alter morphological factors such as TARNs; COND; ASSI; ALT; MPER; Cloa; Clob; CloTO; RUBI and ASSI, among others (Figure 1). The negative control present in the third and fourth quadrants correlates negatively with the variables present in the first and second quadrants (Figure 1). Plant development is closely related to the quantification of gas exchange in leaves comprising liquid CO₂ assimilation, as well as transpiration, stomatic conductance, internal CO₂ concentration in the stomatic chamber, water use efficiency, and instantaneous efficiency of carboxylation, among others [26].

The application of phosphate, nitrogen, and biostimulant fertilization fully influences the biometric, morphological, and physiological variables of mombasa grass, however, the need for further research that can guide the use of products in cultivated fodder is emphasized, presenting the effects promoted in plants and their advantages for Brazilian agriculture.

CONCLUSION

The application of Sulfammo MeTA (SN1; SN3) promotes a positive influence on agronomic, morphogenic, and physiological variables in components 1; 2; 3, and 4.

The joint use of urease inhibitor and Sulfammo MeTA nitrogen source (SN3 - 6kg/ton + ASP4) provides the highest scores in the first four components (PC1; PC2; PC3 and PC4), representing 78.10% of the data.

The negative control presented low scores in all components.

COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

REFERENCES

1. USDA - United States Department of Agriculture. Livestock and poultry: world markets and trade. (Despite HPAI Outbreaks, Global Broiler Meat Production and Trade Forecast to rise in 2018). Approved by the World Agricultural Outlook Board/USDA. Foreign Agricultural Service/USDA. Office of Global Analysis. 2018.
2. ABIEC - Brazilian Association of Meat Exporting Industries. Brazilian Livestock. available at: http://www.abiec.com.br/3_pecuaria.asp Access in 02 Mar. 2018.
3. Oliveira SB, Caione G, Camargo MF, Oliveira ANB, Santana L. Sources of Phosphorus in the Establishment and Productivity of Forages in the Region of High Forest - MT. *Global Science and Technology*, v. 05, n. 01, p. 01–10, 2012
4. Carneiro JSS, Silva PSS, Santos ACM, Freitas GA, Silva RR. Response of Mombasa grass under the effect of phosphorus sources and doses in the formation fertilization. *Journal of Bioenergy and Food Science*, v. 4, n. 1, p. 12-25, 2017.
5. Mendonça VZ, Mello LMM, Andreotti M, Pariz CM, Yano EH, Pereira FCBL. Release of nutrients from straw from forages cocropmyated with corn and succession with soybean. *Brazilian Journal of Soil Sciences*, v.39, p.183-193, 2015.
6. Moreira LM, Martuscello JA, Fonseca DM, Mistura C, Morais RVM, Ribeiro Junior JI. Peresonment, forage accumulation, and bromatological composition of brachiaria grass fertilized with nitrogen. *Brazilian Journal of Animal Science*, vol.38, n.9, pp. 1675-1684, 2009.
7. Primavesi AC, Primavesi O, Corrêa LA, Silva AG, Cantarella H. Nutrients in marandu grass phytomass as a function of nitrogen sources and doses. *Agrotechnical Science*, v.30, p.562-568, 2006.
8. IFA – International fertilizer industry association. Available at: <http://www.fertilizer.org/ifa/Home-Page/STATISTICS/Fertilizer-supply-statistics>; Accessed at 10/08/2019.
9. Cantarella H, Novais RF, Alvarez VVH, Barros NF, Fontes RLF, Cantarutti RB, Neves JCL. (Ed.). Soil fertility. Viçosa, Brazilian Society of Soil Science. p. 1017, 2007.

10. Radel RJ, Gautney J, Peters GE, Urease inhibitor developments. In: Bock BR, Kissel DE. (Ed.) Ammonia volatilization from urea fertilizers. Muscle Shoals, National Fertilizer Development Center. p. 111-136, 1988.
11. Chien SH, Prochnow LI, Cantarella H. Recent developments of fertilizer production and use to improve nutrient efficiency and minimize environmental impacts. *Advances in Agronomy*, v. 102, p. 267-322. 2009.
12. EMBRAPA - Brazilian Agricultural Research Company. National Soil Research Center. Manual of soil analysis methods. 2. ed. Rio de Janeiro: Embrapa, 1997.
13. CFSEMG – Soil Fertility Commission of the State of Minas Gerais. Recommendation for the use of correctives and fertilizers in Minas Gerais - 5th Approximation. Viçosa: UFV, 1999, 359p.
14. Huerta AS, Alvim PT. Leaf area index and its influence on the photosynthetic ability of the café. *Cenicafé*, v. 13, n. 2, p. 75–84, 1962.
15. Gomide MB. et al. Comparison between leaf area determination methods in Mundo Novo and Catuaí coffee trees. *Practical Science*, v. 1, n. 2, p. 118–123, 1977.
16. Hair JFJ. et al. *Análise Multivariada de Dados*. 6. ed. Porto alegre: Bookman, 2009.
17. Team RCRR: A language and environment for statistical computing Vienna, Austria, 2013.
18. Johnson RA, Wichern DW. *Applied multivariate statistical analysis*. 4 ed. Nova Jersey: Prentice-Hall, p. 632, 1998.
19. Silva FD, Pegoraro RF, Martins VM, Kondo MK, Dorasio S, Oliveira GL, Mota MFC. Soil ammonia volatilization after urea doses with urease inhibitors and nitrification in pineapple crop. *Ceres Magazine*, v. 64, n.3, p. 327-335, 2017.
20. Dawar K, Fahad S, Jahangir MMR, Munir I, Alam SS, Khan AS, Danish S. Biochar and urease inhibitor mitigate NH_3 and N_2O emissions and improve wheat yield in a urea fertilized alkaline soil. *Scientific Reports*, v. 11, n. 1, p. 1-11, 2021.
21. Scivittaro WB, Gonçalves DRN, Vale MLC, Ricordi VG. Nitrogen losses by ammonia volatilization and response of irrigated rice to urea application treated with nbpt urease inhibitor. *Rural Science*, v. 40, n. 6, p. 1283-1289, 2010
22. Cantarella H, Trivelin PCO, Contin TLM, Dias FLF, Rossetto R, Marcelino R, Coimbra RB, Quaggio JA. Ammonia volatilization from urease inhibitor-treated urea applied to sugarcane trash blankets. *Scientia Agricola*, v. 65, n. 4, p. 397-401, 2008.
23. Chagas PHM, Gouveia GCC, Costa GGS, Barbosa WFS, Alves AC. Ammonia

volatilization in the pasture is fertilized with nitrogen sources. *Neotropical Agriculture Journal*, v. 4, n. 2, p. 76-80, 2017.

24. Tasca FA, Ernani PR, Rogeri DA, Gatiboni LC, Cassol PC. Soil ammonia volatilization after application of conventional urea or with urease inhibitor. *Brazilian Journal of Soil Science*, v. 35, n. 2, p. 493-502, 2011.

25. Cantarella H, Otto R, Soares JR, Brito Silva AG. Agronomic efficiency of NBPT as a urease inhibitor: A review. *Journal of advanced research*, 13, 19-27, 2018.

26. Taiz L, Zeiger E. *Physiology and plant development Plant diversity*. 6. ed. Porto Alegre: Artmed, 2017.

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