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ORIGINAL RESEARCH PAPER

Organic management of *Urochloa brizantha* cv. Marandu intercropped with leguminous

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Abstract The aim of this work was to evaluate the biomass productivity and chemical-bromatological quality of *Urochloa brizantha* cv. Marandu, intercropped with different leguminous under organic management. The experiment was conducted for 3 years at Brazilian Agricultural Research Corporation — Embrapa Cerrados in Planaltina — DF, Brazil. Two independent experiments were designed. The first one with a previous green manure crop and the second, without green manure crop. After that, were planted in both experimental areas the *Urochloa brizantha* cv. Marandu intercropped with S*tylosanthes* spp. The experimental design was performed in randomized blocks, in the sub-subdivided plot scheme, with 3 replicates and 3 factors. Forage mass production, nutritional value, and protein and carbohydrate fractionation were evaluated in trials. *Urochloa* presented better quality in the organic system, with the lower lignin content (3.9%) and the reduction of low availability nitrogenous compounds. *Stylosanthes* spp. presented better chemical-bromatological quality with green fertilization in the form of organic fertilization, due to the significant reduction of fibers and lignin, a result that promoted improvements in digestibility. Both *Urochloa* and *Stylosanthes* presented better nutritional quality in the system with green fertilization and organic form of fertilization.

Keywords Healthy ecosystems · Legume · Organic fertilizer · Sustainability

Introduction

Brazilian livestock production is concentrated in the use of pasture, occupying a total area of 158,622,704 hectares, of which 46,847,430 hectares are of natural pastures, 11,821,006 of degraded planted pastures, and 99,954,268 hectares of pasture planted in good condition (IBGE 2017). Pastures of *Urochloa brizantha* cv. Marandu, which is considered the most used forage in Brazil, are responsible for about 40% of the sales of forage seeds and cultivated in approximately 70 million hectares (Freire et al. 2016).

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Pastures have great relevance in the production systems because that are characterized as the most economical and practical way of producing and supplying feed to cattle (Boval and Dixon 2012; Dias Filho 2014), in addition to providing relevant ecosystem services such as water abstraction, biodiversity reserves, native pastures, and, potentially, carbon sequestration in the soil, reducing greenhouse gas emissions (Boval and Dixon 2012).

However, one of the greatest challenges found in this production system is the fact that most of these pastures are in some degree of degradation (Dias Filho 2017). Another great problem is the lack of crop rotation, which may favor the appearance of pests and/or agricultural diseases and also the emergence of invasive plants (Pertot et al. 2017), and consequent increase in the use of agrochemicals that may occasionally contaminate the environment (Zimdahl 2018). The use of chemical fertilizers in the process of renovation or recovery of degraded areas is very expensive (Heneghan et al. 2008; Macedo et al. 2013). In general, they are used incorrectly causing edaphoclimatic damage such as acidification, eutrophication, and other impacts to air, water, and soil (Eggleston et al. 2006), and most chemical fertilizers originate from non-renewable mineral resources (Steinbach and Wellmer 2010).

One of the alternatives to overcome the problem of pasture degradation is the renewal or recovery of these using the consortium of legumes and grasses (Lüscher et al. 2014) and green manures, which it had as objective the incorporation of organic matter and nitrogen in the soil (Alcântara et al. 2000). In addition to the use of these alternatives falling within the organic production, which is a production model, that seeks the sustainability of the production system. Intercropping fodder legumes in pastures contribute to N uptake by the grasses, increasing pasture support. Additionally, N fixed by the fodder legumes may improve the dietary quality and growth of animals (Lüscher et al. 2014; Pirhofer-Walzl et al. 2012). Also, during the dry season, when the nutritive value of pastures decreases, animals are better nourished in pastures intercropped with fodder legumes (Azevedo Júnior et al. 2012). All these strategies combined make it possible to intensify agricultural activities in a sustainable manner (Kohmann 2017; Gianezini et al. 2019).

In organic management systems, the use of alternative regional fertilizers, such as the agrominerals and cattle manure, is strategic for pasture crops grown in low fertility soils, such as Cerrado soils (Brasil 2011; Soares et al. 2014, 2018). The cattle manure may be used in the fertilization of fodder replacing in parts the inorganic fertilizer (Castro et al. 2016). Amending the acidity and fertility of these soils is paramount for pasture growth and increase in forage nutrition levels in conventional or organic management systems.

Another hand, to maximize the use of the nutrients by the animals the Cornell Net Carbohydrate and Protein System (CNCPS) is a nutritional model that evaluates the environmental and nutritional resources available in an animal production system and enables the formulation of diets that closely match the predicted animal requirements (Van Amburgh et al. 2015), strategical too for forages management under organic management.

In general, the analyses show that organic animal production systems are economically viable, with a diversity of productive arrangements and better gains, because they present differentiated products, besides contributing to plant production through natural fertilizers (Soares et al. 2018; Figueiredo and Soares 2012).

Although organic animal production is present worldwide, according to Willer and Lernoud (2019), the available statistics on the number of organic animals are still incomplete and do not allow an overview of the sector. However, it is possible to observe from the available information the advance of organic animal production mainly in European countries. Thus, the largest participations are sheep and cattle, because they are extensive productions that become easier for organic conversion (Willer and Lernoud 2019).

Sustainable and bioeconomic livestock technologies are required for organic animal production systems, and the growth response of forage from tropical species to legally accepted alternative fertilizers for organic systems is still unknown (Figueiredo and Soares 2012; Soares et al. 2016). In this trial, the aim was to evaluate the biomass productivity and nutritional composition of *Urochloa brizantha* cv. Marandu in association with *Stylosanthes* spp. and other Legumes under organic management.

Material and methods

Study area

The experiment was conducted at Brazilian Agricultural Research Corporation — Embrapa Cerrados in Planaltina — DF, Brazil (15° 35′ 30″ S; 47° 42′ 30″ W), with an average elevation of 1007 m, climate type is AW, according to Köppen; annual average precipitation of 1460 mm (Table 1).

The soil was classified as a Latossolo Vermelho distrófico (dos Santos et al. 2018), i.e., an Ustox Acrustox Oxisol (Soil Survey Staff 2015), with a low fertility, a clay texture, and the following chemical characteristics at the 0–20 cm depth: pH = 5,8; Al = 0.04 cmolc dm-^{3}; Ca + Mg = 1.54 cmolc dm-^{3}, P = 3.61 mg dm-^{3} e K = 47 mg dm-^{3}. The area had been fallow for 10 years, with no recent cultivation. It was chosen for the experiment because the low natural fertility would be responsive to the evaluated fertilizers. There was only pasture composed of grasses (genera *Urochloa* and *Panicum*) in different stages of degradation.

Experimental trials

Two independent experiments were designed, the first one with a previous green manure crop, which was incorporated into the soil before experiment implementation, and the second, without a green manure crop. After that, experiments were installed with *Urochloa brizantha* cv. Marandu consortium with S*tylosanthes* spp. in plots of 10×12 m. The experimental design was performed in randomized blocks, in the sub-subdivided plot scheme, with 3 replicates and 3 factors. In the plots, three forms of soil management being them conventional (CON), organic (ORG), and untreated (UNT) were performed in the sub-plots, two planting operations that went without and with Guandu bean (*Cajanus cajan*) and in the sub-sub plot (with and without mycorrhiza) were carried out.

Table 1 Air temperature, and annual precipitation, Planaltina, DF

Year	Air temperature (°C)			Precipitation (mm)
	Max	Min	Average	
2013	28.2	16.4	21.6	1318.1
2014	28.7	15.9	21.5	1350.3
2015	27.8	16.0	21.2	1415.3

Two adjacent areas of 2160 m^{2} each were selected. Two Mg of limestone ha^{-1} and 1 Mg gypsum ha^{-1} were applied in both areas. Then, after 3 months, 30 kg ha^{-1} *Crotalaria juncea* inoculated seeds (100 g of inoculant for each 10 kg of seeds) were sowed. After 84 days of growth, 705 kg dry weight ha^{-1} of Crotalaria was incorporated into the soil. Crotalaria shoot analysis showed the following results: 24 g kg^{-1} N; 1.48 g kg^{-1} P; 15.5 g kg^{-1} K; 8.1 mg kg^{-1} Ca; 2.3 mg kg^{-1} Mg, representing 78, 4.5, 50.5, 26.4 and 7.5 kg ha^{-1}, respectively for N, P, K, Ca, and Mg.

The different treatments and pre-treatments, correction, fertilization, and planting beyond the dates when the operations were performed in the experimental area can be observed in Table 2.

After Crotalaria was incorporated in the area with green manure, the amounts of fertilizer applied in the organic treatment were 7 Mg ha^{-1} of chicken manure (1.5% N), 2 Mg ha^{-1} of thermopotassium (6% K_2O), and 1 Mg ha^{-1} of thermophosphate (12% P_2O_5) as sources of N, K, and P. The conventional treatment were 200 kg ha^{-1} of potassium chloride (60% K_2O), 260 kg ha^{-1} of triple superphosphate (46% P_2O_5), and 217 kg ha^{-1} of urea (46% N). The total supply of nutrients applied to each treatment (conventional or organic) was equivalent to 100 kg N ha^{-1}; 120 kg P_2O ha^{-1}, and 120 kg K_2O ha^{-1}. For the untreated plots was not performed any fertilization. The control did not receive any fertilizer, except lime and gypsum.

After fertilized both experiments were sowed with mixed seeds of legumes and grass seeds. Three kg ha^{-1} of *Stylosanthes* spp., and 2.5 kg ha^{-1} of *Urochloa brizantha* cv. Marandu was used. The legumes were *Stylosanthes guianensis* cv. Mineirão (20%), *Stylosanthes capitata-macrocephala* cv. Campo Grande (20%) and *Stylosanthes guianensis* cv. Bela (60%) intercalated with rows (30 seeds/linear meters) of pigeon pea (*Cajanus cajan* cv. Mandarim) in different plots, with and without rows of pigeon pea. Leguminous seeds and grasses were inoculated with mycorrhizal fungi and *Acetobacter diazotrophic*, respectively, except *Stylosanthes* spp.

Biomass production and chemical composition

The experiment was evaluated every 6 months, for 3 years (2013–2015) in a row. The cuttings were made at 10 cm from ground level, within a square of 1 m2 thrown randomly in three different points of each plot. After that, the produced biomass was removed from the area. The cuts were performed in different data of the dry and rainy seasons of each year: 26/03/2013; 16/07/2013; 15/01/2014; 25/07/2014; 17/02/2015; and 02/08/2015. Collected shoot samples were separated by grass or legume and weighed in the field. The percentage of fresh weight of grass and legume plants was then calculated. Samples were subjected to drying in an oven at 65 °C for 72 h. Dried samples were weighed and analyzed chemically.

Chemical analysis was performed according to (AOAC 2000). DM (Association of Official Analytical Chemistry (AOAC) 950.15), ash (AOAC 942.05), ether extract (EE, AOAC 920.39), CP ($CP = N \times 6.25$; AOAC 984.13) and lignin (AOAC 973.18) according to the methods described by AOAC (2000). NDF was analyzed using α-amylase without the addition of sodium sulfite to the detergent (TE-149 fiber analyzer; Tecnal Equipments for Laboratory Inc., Piracicaba, Brazil). ADF was determined as described by Van Soest et al. (1991). Non-fiber carbohydrates (NFC) concentrations were estimated according to Hall (2000) where $NFC = 100 - [\%CP + \%EE + \%ash + \%NDF]$.

In vitro digestibility of dry matter

The IVDMD of the diets was then determined using the artificial rumen (DaisyII Fermenter®, Ankom) with ruminal liquid collected in fasting from a donor animal, adapted for 10 days to green forage (McDougall 1948; Tilley and Terry 1963; Goering and Van Soest 1970). Then 0.5 g of the sample was packed in TNT-100 g/m bags, cut and sealed to a size of 5.0 × 5.0 cm, as were determined according to the method proposed by Casali et al. (2008), where two sachets without a sample (white) and those with samples were placed in the jars, evenly distributed. The incubation was stopped after 48 h, and pepsin and hydrochloric acid were added to stop fermentation and microbial enzymatic activity to simulate the second part of the digestibility for another 48 h. At the end of the second stage, the bags were washed with running water until the residue was completely removed from the digestibility and transferred to a drying oven at 105 °C for 12 h and the in vitro dry matter digestibility was determined by weight difference. The IVDMD was then calculated using the formula: $IVDMD = [A - (B - C) * 100]/A$. At where: A = initial dry matter = sample weight × 2DM / 100; B = residual dry matter = crucible weight with dry residue—crucible weight- C; C = this is the white, consists of empty sachets.

Protein and carbohydrate fractionation

Protein fractionation was performed according to Licitra et al. (1996), that is, fraction A (NNP) was obtained by the difference between total nitrogen and insoluble nitrogen in trichloroacetic acid. Total insoluble nitrogen was determined from the treatment of 0.5 g of the sample with borate-phosphate buffer. Total soluble nitrogen was obtained by the difference between total nitrogen, minus insoluble nitrogen in the borate-phosphate buffer. The B1 fraction was determined by the difference between the total soluble nitrogen fraction minus A fraction. The B3 fraction was calculated by the difference between neutral detergent insoluble nitrogen (NDIP) and acid detergent insoluble nitrogen (ADIP), determined by boiling 0.5 g of the sample, with a neutral and acidic detergent solution for 1 h, respectively, with analysis of residues also for nitrogen. The C fraction was considered as acid detergent insoluble nitrogen (ADIN), and the B2 fraction, determined by the difference between 100 and fractions A, B1, B3, and C, as a percentage of the protein.

Total carbohydrates (TC) and non-fibrous (CNF) were determined according to Sniffen et al. (1992), by the expressions $TC = 100 - (\%\ CP + \%\ FAT + \%\ ASH)$, and $CNF = 100 - (\%\ CP + \%\ FAT + \%\ NDFcp + \%ASH)$, where NDFcp is equivalent to the neutral detergent fiber corrected for ashes and proteins. The B2 fraction was calculated by the difference between FDNcp – fraction C (Sniffen et al. 1992) and C, by the percentage of lignin multiplied by 2.4 (Sniffen et al. 1992).

Statistical analyzes

Multivariate analyses were performed on bromatological, fractionation, digestibility, and production data, to verify chemical quality differentiation patterns in forages under different fertilization management. Data were tested for normality of residues and homogeneity of variances using PROC UNIVARIATE (SAS 2012). Also, PROC MIXED of SAS, version 9.0, was used fc model:

Table 2 Management and implementation of the experimental area

Management	Date	CON	ORG	UNT
With green fertilizer				
Correction	08/05/12	2 Mg of limestone ha^{-1} and 1 Mg gypsum ha^{-1}		
Crotalaria Seeding	11/06/12	30 kg ha^{-1} *Crotalaria juncea* inoculated seeds		
Crotalaria incorporation	01/29/13	705 kg dry matter ha^{-1} of Crotalaria was incorporated into the soil		
Fertilization	02/01/13	Urea, 217 kg ha^{-1} (46% N); Potassium chloride, 200 kg ha^{-1}(60% K2O); Triplesuperphosphate, 260 kg ha^{-1} (46% P2O5)	Chicken manure, 7 Mg ha^{-1}(1.5% N); Thermopotassium, 2 Mg ha^{-1}(6% K_2O); Thermophosphate, 1 Mg ha^{-1} (12% P2O5)	-
Pasture Seeding	02/05/13	3 kg ha^{-1}*Stylosanthes* spp. mixed 2.5 kg ha^{-1} of *Urochloa brizantha* cv. Marandu *Cajanus cajan* cv. Mandarim (30 seeds/linear meters)		
Pasture Harvest	2013–2015	The cuts were performed in different data of the dry and rainy season of each year		
Without green fertilizer				
Correction	08/05/12	Two Mg of limestone ha^{-1} and 1 Mg gypsum ha^{-1}		
Crotalaria Seeding	-	-	-	-
Crotalaria incorporation	-	-	-	-
Fertilization	02/01/13	Urea, 217 kg ha^{-1} (46% N); Potassium chloride, 200 kg ha^{-1} (60% K_2O); Triplesuperphosphate, 260 kg ha^{-1} (46% P_2O_5)	Chicken manure, 7 Mg ha^{-1} (1.5% N); Thermopotassium, 2 Mg ha^{-1} (6% K_2O); Thermophosphate, 1 Mg ha^{-1} (12% P_2O_5)	-
Pasture Seeding	02/05/13	3 kg ha^{-1} *Stylosanthes* spp. mixed 2.5 kg ha^{-1} of *Urochloa brizantha* cv. Marandu *Cajanus cajan* cv. Mandarim (30 seeds/linear meters)		
Pasture Harvest	2013–2015	The cuts were performed in different data of the dry and rainy season of each year		

$$\begin{aligned} Y = {} & B_{(i)} + S_{(j)} + A_{(k)} + M_{(l)} + Gm + S_{(i)} * A(k) + S(j) * M(l) \\ & + S(j) * G(m) + A(k)? * M(l) + A(k) * G(m) + G(m) * M(l) + e_{ijklm} \end{aligned}$$

Effects:

B_i = Block Effect (i = 1 a 3)

$S_{(j)}$ – System Effect (j = 1 a 3)

$A_{(k)}$ – Fertilization Effect (k = 1 a 2)

$M_{(l)}$ – Mycorrhiza effect (l = 1 a 2)

$G_{(m)}$ – Guandu effect (m = 1 a 2)

Interactions = $S_{(j)}*A_{(k)}$ interaction effect between green manure system and manure forms; $S_{(j)}*M_{(l)}$ interaction effect between fertilization system and mycorrhiza; $S_{(j)}*G_{(m)}$interaction effect between green and guandu manure system; $A_{(k)}*M_{(l)}$ interaction effect between forms of fertilization and mycorrhiza; $A_{(K)}*G_{(m)}$ interaction effect between manure and guandu forms; $M_{(l)}*G_{(m)}$ interaction effect between mycorrhiza and guandu; $S_{(J)}*A_{(K)}*M_{(l)}$ interaction effect between green manure system, manure forms, and mycorrhiza; $S_{(J)}*A_{(K)*}G_{(m)}$ interaction effect between green manure system, manure forms, and guandu; $S_{(J)}*M_{(l)}*G_{(m)}$ interaction effect between green manure system, mycorrhiza, and guandu; $A_{(K)}*M_{(l)}*G_{(m)}$ interaction effect between forms of fertilization, mycorrhiza, and gunadu; $S_{(J)}*A_{(K)}*M_{(l)}*G_{(m)}$ interaction effect between green manure system, manure forms, mycorrhiza, and guandu.

Year and season were considered as random effects in the model. The means were compared using the procedure LSMEANS and tested with adjusted Tukey test of PROC MIXED.

Results

The results found were not evaluated for triple and quadruple interactions.

The results of dry matter (DM) production and the forage and legume percentage submitted to the different fertilization forms from the six cuts performed in the 3 years of evaluation are shown in Table 3.

Table 3 Dry matter yield of *Urochloa brizantha* cv. Marandu, *Stylosanthes* spp. and percentage of forage and legume in consortium submitted to different forms of fertilization

Item	With green fertilizer			Without green fertilizer			SEM	P-value		
	CONV	ORG	UNT	CONV	ORG	UNT		System	Form	Interaction
Kg ha^{-1}										
UC	6642.88	6758.00	6699.27	6982.46	6628.04	5764.72	118.79	0.08	0.28	0.06
ST	767.79^{bA}	894.53^{abA}	534.25^{cB}	804.52^{aA}	1032.36^{aA}	986.4^{aA}	35.18	<0.01	<0.01	<0.01
CS	7410.66	7652.53	7233.52^{A}	7786.98	7660.4	6751.11^{B}	129.53	0.03	0.88	0.32
%										
UC	0.89^{aA}	0.88^{aA}	0.92^{aA}	0.91^{aA}	0.87^{bA}	0.84^{bB}	<0.01	0.01	<0.01	<0.01
ST	0.11^{aA}	0.12^{aA}	0.08^{bB}	0.09^{bA}	0.13^{aA}	0.16^{aA}	<0.01	<0.01	0.15	<0.01

The averages followed by similar characters do not differ by 5% between conventional (CON), organic (ORG) and untreated (UNT) forms of fertilization. The followers of equal letters cannot differ by 5% between systems of green fertilizer and without green fertilizer. *UC Urochloa*; *ST Stylosanthes*; *CS* consortium of *Urochloa brizantha* cv. Marandu and *Stylosanthes* spp

It can be observed that there was no interaction ($P>0.05$) between the fertilization systems (with green fertilization and without green fertilization) and conventional (CON), organic (ORG), and untreated (UNT) fertilization forms for the variable DM productivity (kg.ha^{-1}) yields of *Urochloa brizantha* cv. Marandu and the consortium between *Urochloa* and *Stylosanthes* spp.

The dry matter yield of *Stylosanthes* spp. and *Urochloa brizantha* cv. Marandu in consortium presented significant differences ($P<0.05$) in the yield of DM (kg.ha^{-1}) of the *Urochloa brizantha* cv. Marandu consortium with *Stylosanthes* spp. was observed for a year and climatic season. The dry matter yield of *Stylosanthes* spp. (kg.ha^{-1}) it presented an interaction ($P<0.05$) between the forms of fertilization and the presence or not of the green fertilization.

For the DM of *Urochloa brizantha* cv. Marandu was observed to have a significant effect ($P<0.05$) about the year and season. There was no effect ($P>0.05$) on DM production about fertilization forms with and without green fertilization.

Due to the importance of carbohydrates and proteins present in forage for ruminant nutrition, the fractions of these components were analyzed in organic and conventional fertilization models to compare their effect on forage composition. The results of these analyzes can be seen in Table 4.

All carbohydrate fractions *Urochloa brizantha* cv. Marandu presented interaction ($P<0.05$) between green fertilization and conventional fertilization. There was no statistical difference ($P>0.05$) for TC and NFC parameters, that is, regardless of the form of fertilization, the proportion of TC and NFC were the same with the use of green fertilizer.

Carbohydrate fractions in organic and conventional showed interaction ($P<0.05$) between green fertilization and fertilization forms (Table 4). There was no statistical difference ($P>0.05$) for the CHT and NEC parameters, that is, regardless of the form of fertilization, the proportion of CHT and NEC were the same with green fertilizer. When analyzing the system without green fertilization, it was verified that there was a difference for NFC where the organic fertilization form.

There were no significant differences ($P<0.05$) for ASH, OM, FAT, NDIN, and ADIN for with green fertilizer and without green fertilizer systems regardless of the form of fertilization either CON or ORG. There was interaction ($P<0.05$) with green fertilizing versus fertilization forms for CP content, with emphasis on the higher value of ORG treatment in plots with fertilizer (Table 5).

The averages followed by similar characters do not differ by 5% between conventional (CON), organic (ORG), and untreated (UNT) forms of fertilization. The followers of equal letters cannot differ by 5% between systems of green fertilizer and without green fertilizer.

The levels of NDF, NDFap, and ADF also experienced interaction ($P<0.05$) between green fertilizing and the fertilization forms.

There was interaction ($P<0.05$) between green fertilizing versus the fertilization forms for the LIG content, where the values were lower and consequently better for green fertilizer in the organic system.

Table 4 Fractionation of nitrogen compounds and the fractionation of carbohydrates of *Stylosanthes* spp. and *Urochloa brizantha* cv. Marandu intercropped with *Stylosanthes* spp. in the treatments

Item (%)	With green fertilizer			Without green fertilizer			SEM	P-value		
	CON	ORG	UNT	CON	ORG	UNT		System	Form	Interaction
Stylosanthes spp.										
FA1	36.38^{aB}	37.14^{aA}	33.97^{cB}	41.64^{aA}	38.36^{bA}	41.37^{aA}	0.32	< 0.01	< 0.01	< 0.01
FB12	7.23^{aA}	6.43^{aB}	6.93^{aA}	5.52^{bA}	8.69^{aA}	8.06^{aA}	0.19	0.02	< 0.01	< 0.01
FB23	23.43^{bA}	27.00^{aA}	26.45^{aA}	22.95^{aA}	17.44^{bB}	19.88^{bB}	0.34	< 0.01	0.16	< 0.01
FB34	21.35^{aA}	18.73^{bB}	20.99^{aA}	19.19^{bB}	22.92^{aA}	19.69^{bA}	0.20	0.33	0.15	< 0.01
FC5	11.6	10.69	11.65	10.68	12.57	10.98	0.10	0.55	0.33	0.56
TC6	81.16^{aA}	80.14^{aA}	81.40^{aA}	79.58^{aB}	80.09^{aA}	79.43^{aB}	0.14	< 0.01	0.51	< 0.01
NFC7	24.34	25.3	24.09	23.23	21.15	21.33	0.24	< 0.01	< 0.01	0.31
A + B1	32.33	31.51	29.54	29.19	26.38	26.82	0.28	< 0.01	< 0.01	0.06
B2	47.59	49.45	50.27	51.70	53.77	53.94	0.39	< 0.01	< 0.01	0.86
C	19.99^{aA}	19.03^{aB}	20.20^{aA}	19.88^{aA}	20.73^{aA}	19.40^{aA}	0.19	0.18	0.85	< 0.01
Urochloa brizantha cv. Marandu intercropped with *Stylosanthes* spp.										
FA1	38.32^{aA}	35.62^{bA}	30.1^{cB}	41.88^{aA}	32.46^{bA}	35.94^{bA}	0.45	< 0.01	< 0.01	< 0.01
FB12	5.82^{bA}	12.93^{aA}	7.27^{bB}	7.21^{bA}	9.29^{aB}	9.6^{aA}	0.21	0.90	< 0.01	< 0.01
FB23	20.84^{b}	21.98^{b}	27.01^{a}	19.13^{b}	22.19^{b}	26.45^{a}	0.38	0.13	< 0.01	0.22
FB34	17.4^{aA}	11.94^{bB}	19.01^{aA}	15.06^{aB}	17.1^{aA}	10.85^{bB}	0.27	< 0.01	< 0.01	< 0.01
FC5	17.62^{aA}	17.52^{aB}	16.6^{aA}	16.71^{bA}	18.95^{aA}	17.15^{bA}	0.12	0.06	< 0.01	< 0.01
TC6	82.49^{aB}	82.18^{aB}	82.37^{aA}	83.82^{aA}	83.84^{aA}	82.37^{aA}	0.10	< 0.01	< 0.01	< 0.01
NFC7	14.67^{aA}	14.24^{aB}	14.8^{aA}	14.89^{bA}	17.21^{aA}	15.75^{bA}	0.15	< 0.01	0.01	< 0.01
A + B1	17.71^{aA}	17.31^{aB}	17.95^{aA}	17.71^{bA}	20.49^{aA}	19.07^{bA}	0.17	< 0.01	< 0.01	< 0.01
B2	70.96^{bA}	73.32^{aA}	70.22^{bA}	71.48^{aA}	69.2^{bB}	69.67^{bA}	0.20	< 0.01	< 0.01	< 0.01
C	11.33^{aA}	9.37^{bA}	11.83^{aA}	10.8^{aA}	10.3^{aA}	11.25^{aA}	0.11	0.77	< 0.01	< 0.01

FA1 Fraction A, *FB12* Fraction B1, *FB23* Fraction B2, *FB34* Fraction B3, *C5* Fraction C. The averages followed by similar characters do not differ by 5% between conventional (CON), organic (ORG), and untreated (UNT) forms of fertilization. The followers of equal letters cannot differ by 5% between systems of green fertilizer and without green fertilizer. *TC6* total carbohydrate; *NFC7* non-fibrous carbohydrates. The averages followed by similar characters do not differ by 5% between conventional (CON), organic (ORG), and untreated (UNT) forms of fertilization. The followers of equal letters cannot differ by 5% between systems of green fertilizer and without green fertilizer

The dry matter yield of *Urochloa and Stylosanthes* consortium percentages affected ($P < 0.05$) in the fertilization forms and green fertilization. For *Urochloa* the percentage with green fertilization there was no difference ($P > 0.05$) between the fertilization forms different from the one observed without green fertilization in which the conventional one presented higher establishment value in the plots. It was observed that there was an interaction effect ($P < 0.05$) between green and Guandu bean fertilization for dry matter production of *Stylosanthes* spp.

It was observed that there was an interaction effect ($P < 0.05$) between green fertilization and mycorrhizal fungi for dry matter production of *Stylosanthes* spp.

For the analysis of the *Stylosanthes* spp., there was interaction ($P < 0.05$) between green fertilization for the variables DM, CP, NDF, ADF, CIDN, LIG, FAT, and IVDMD. However, there were no significant differences for ASH, OM, FAT, NDIP, ADIP for systems using green fertilizer and without green fertilization for both the organic and conventional models. There was also interaction ($P < 0.05$) with green fertilizer versus fertilizer for the DM content.

Analyzing the in vitro digestibility of *Urochloa* dry matter (Table 5) about green fertilizer and fertilization forms, the presence of interaction ($P < 0.05$) was verified for this parameter where UNT presented a higher result with green fertilizing.

Table 5 Chemical composition of *Stylosanthes* spp. and *Urochloa brizantha* cv. Marandu intercropped with *Stylosanthes* spp

Item (%)	With green fertilizer			Without green fertilizer			SEM	P value		
	CON	ORG	UNT	CON	ORG	UNT		System	Form	Interaction
Stylosanthes spp.										
DM	45.96^{aA}	43.78^{aB}	44.57^{aB}	46.85^{bA}	51.01^{aA}	52.27^{aA}	0.55	<0.01	0.1205	<0.01
ASH	5.91	6.04	6.05	5.48	5.49	5.6	0.03	<0.01	0.11	0.56
OM	94.09	93.95	93.95	94.52	94.51	94.39	0.03	<0.01	0.11	0.56
CP	12.79^{aA}	12.77^{aA}	11.33^{bB}	13.85^{aA}	12.78^{aA}	13.38^{aA}	0.13	<0.01	<0.01	<0.01
NDF	60.17^{bA}	59.88^{bB}	62.65^{aA}	61.5^{bA}	65.09^{aA}	63.51^{bA}	0.24	<0.01	<0.01	<0.01
ADF	42.18^{bB}	43.27^{bB}	44.32^{aA}	43.79^{bA}	45.93^{aA}	45.35^{aA}	0.17	<0.01	<0.01	0.03
CIDN	55.11^{bB}	54.85^{bB}	57.39^{aA}	56.34^{bA}	59.62^{aA}	58.18^{aA}	0.22	<0.01	<0.01	<0.01
LIG	8.33^{aA}	7.93^{aB}	8.42^{aA}	8.28^{aA}	8.64^{aA}	8.08^{aA}	0.07	0.18	0.85	<0.01
FAT	2.22^{bB}	3.11^{aA}	3.13^{aB}	3.36^{bA}	3.15^{bA}	4.12^{aA}	0.07	<0.01	<0.01	<0.01
NIDN	0.67^{b}	0.67^{b}	0.7^{a}	0.69^{ab}	0.73^{a}	0.71^{a}	<0.01	<0.01	<0.01	<0.01
ADIN	0.24	0.24	0.25	0.24	0.26	0.25	<0.01	<0.01	<0.01	0.03
IVDMD	57.37^{aA}	54.24^{bA}	52.05^{cB}	56.04^{aA}	53.98^{bA}	56.64^{aA}	0.24	<0.01	<0.01	<0.01
Urochloa brizantha cv. Marandu intercropped with *Stylosanthes* spp.										
DM	47.5^{aA}	35.21^{bA}	48.63^{aA}	40.17^{aB}	36.66^{aA}	42.56^{aB}	0.50	<0.01	<0.01	<0.01
ASH	8.99	9.67	9.48	8.89	9.28	9.42	0.05	0.04	<0.01	0.27
OM	91.01	90.33	90.52	91.11	90.72	90.58	0.05	0.04	<0.01	0.27
CP	6.17^{aA}	6.23^{aA}	6.13^{aB}	6.19^{aA}	5.48^{bB}	6.71^{aA}	0.05	0.52	<0.01	<0.01
NDF	71.96^{aB}	72.09^{aA}	71.71^{aA}	73.15^{aA}	70.66^{bB}	70.67^{bA}	0.12	0.04	<0.01	<0.01
ADF	35.92^{aB}	35.33^{bA}	34.99^{bA}	37.25^{aA}	34.7^{cA}	35.71^{bA}	0.09	<0.01	<0.01	<0.01
CIDN	67.82^{aA}	67.94^{aA}	67.57^{aA}	68.94^{aA}	66.63^{bB}	66.62^{bA}	0.12	0.06	<0.01	<0.01
LIG	4.72^{aA}	3.9^{bB}	4.93^{aA}	4.5^{aA}	4.57^{aA}	4.69^{aA}	0.05	0.45	<0.01	<0.01
FAT	2.45	2.06	2.19	1.66	1.58	1.76	0.04	<0.01	0.04	0.12
NIDN	0.34	0.34	0.33	0.34	0.33	0.33	<0.01	0.67	<0.01	<0.01
ADIN	0.18	0.17	0.17	0.19	0.17	0.18	<0.01	<0.01	<0.01	<0.01
IVDMD	51.6^{bB}	54.1^{aB}	55.91^{aA}	62.95^{aA}	62.6^{aA}	56.3^{bA}	0.48	<0.01	<0.01	<0.01

DM dry matter, *ASH* ash, *OM* organic matter, *CP* crude protein,, *NDF* neutral detergent fiber, *ADF* acid detergent fiber, *CIND* corrected neutral detergent fiber, *FAT* fats, *LIG* lignin, *NIDN* neutral detergent insoluble nitrogen, *ADIN* acid detergent insoluble nitrogen, *IVDMD* in vitro dry matter digestibility

It was observed that the plots with Guandu bean rows were significantly higher ($P<0.05$) in terms of DM when inoculated with mycorrhizal fungi about plots without Guandu, even with inoculation.

Discussion

Comparing the effect of green fertilization for the consortium, the portion without treatment and the green fertilization had a production of 6758.00 kg.ha^{-1} value lower than that of the unprocessed portion and with green fertilization, which reached a production of 7233.52 kg.ha^{-1}. Thus, in the UNT control treatment, it can be concluded that the production was 6.67% lower than the production with green fertilizer. We can infer that the presence of green fertilizer, even without the application of no fertilization, alters the soil characteristics favoring the production corroborating with results found by (Garcia et al. 2008; Souza et al. 2016; Faissal et al. 2017).

The average values of the DM yield of the *Urochloa brizantha* cv. Marandu consortium with *Stylosanthes* spp. presented values for the ORG management with 6905.74 kg.ha^{-1} and 8407.18 kg.ha^{-1}, respectively, and for the conventional management 6568.0 kg.ha^{-1} for the dry period and 8635.64 kg.ha^{-1} for the rainy. The DM production in the second year of evaluation in the dry season and the rainy were lower for both CON fertilization (6257.69 kg.ha^{-1} and 8059.16 kg.

ha^{-1}) and ORG (6730.62 kg.ha^{-1} and 8013.25 kg. ha^{-1}). This behavior was inversely related to the third year, which presented the highest values of productivity but was related to the use of fertilization with the natural phosphorus and potassium sources, which present slower release (Stockdale et al. 2001; Mikkelsen 2007).

Regarding the DM productivity (kg.ha^{-1}) of *Stylosanthes* spp. it presented higher average values for ORG management with 894.53 kg.ha^{-1} followed by CON management 767.79 kg.ha^{-1} and lastly the UNT with 534.25 kg.ha^{-1}, certainly the (14.17%) compared to conventional and UNT (40.28%) which, as discussed above, was already expected for organic management (Stockdale et al. 2001; Mikkelsen 2007). The response of the plants in this form of fertilization becomes smaller, but more constant Stockdale et al. (2001); Stockdale et al. (2002) which may have provided an increase in DM production due to the increase of organic matter of the crotalaria used as green fertilizer in the organic management and that were incorporated in the pasture planting. This production of *Stylosanthes* confirms a greater capacity of this legume to develop in soils with low nutrient availability or low fertility Miranda et al. (2015).

About the year and season, the DM of *Urochloa brizantha* cv. Marandu was observed an increase in DM yield in the period of the waters due to the better climatic conditions such as the temperature and the rainfall (Doorenbos and Kassam 1979; Valle et al. 2000; Aroeira et al. 2005; Lu et al. 2017). The lowest DM production value was 5764.72 kg.ha^{-1} for the UNT plot, which is more than sufficient to meet the consumption demand of a 500 kg animal is around 3% of the weight live or 1500 kg (Dickerson 1970).

Among the forms of fertilization, the lowest values of total carbohydrates were observed for the forms of conventional and organic fertilization with green fertilizer, a result that can be justified by the lower DM content in these forms of fertilization. It can also be observed that although the organic matter presented lower levels of DM than the conventional form of fertilization, it was verified that for the parameter TC, the organic fertilization was better than the conventional fertilization. When analyzing the system without green manure, it was verified that the form of organic fertilization stands out (17.21%) about the others.

Ruminants are herbivorous animals whose main characteristic is the transformation of structural carbohydrates into meat, milk, and wool. In a study carried out by Silva and Silva (2013), the authors concluded that the nutrition and feeding of ruminants are areas of knowledge that require different scientific information for the development of techniques to adequately nourish the animals of zootechnical interest among them the analysis of the fractionation of carbohydrates and proteins. In this sense, to resort to the studies of zootechnical bromatology is of extreme importance, because allows us to know the number of nutrients that the food can offer to satisfy the needs of the animal. In formulating diets the carbohydrates are the main constituents of forages, corresponding to 50 to 80% of DM of forages and cereals (Silva and Silva 2013). When analyzing the system without green fertilization, it was verified that NFC stands out (17.21%) in organic fertilization about the others.

Between treatments, the values of CP showed higher value of ORG treatment in plots with fertilizer (6.23%); in contrast to the treatment without green fertilizer (5.48%), result was already expected, because the green fertilizer improves soil quality and favor the availability and use of nutrients by grasses (Franco et al. 2003).

Velásquez et al. (2010), in a study evaluating the same Urochloa species with a 42 day of growth, showed values of 14.08% CP. Mari (2003) in a study also evaluating the same species, with a cut age of 90 days found values of 8.9% CP. Van Soest (1994) described that with the growth of the forages occur the increase of the thickness of the cell wall and the raising the values of NDF that can decrease the levels of the organic molecules reducing the concentration of nitrogen compounds.

The NDF results show that with the green fertilizer the CON system was favored (71.96%) lower than that of the green fertilizer (73.15%). As for the fertilization form ORG, the lowest values for this variable are the best results, however, as the NDF is composed of cellulose and lignin (LIG) which are considered indigestible compounds and also by hemicellulose that has a medium digestibility (Allinson and Osbourn 1970; Fahey and Jung 1983; Susmel and Stefanon 1993; Bravo et al. 1994; Van Soest 1994; Bravo and Saura-Calixto 1998). Therefore it is observed that the presence of was lower (3.9%) in the system with green fertilizer than in the absence of green fertilizer (4.57%), showing that the presence of green fertilizer in the form of organic fertilization favors the increase of nutrients with greater capacity of use by ruminants.

Also, when the determination of the hemicellulose values by the NDF and ADF values is performed, the following values 36.04%, 36.76%, and 36.72% for CON, ORG, and UNT, respectively, with green fertilizer in counterpart in the system without green fertilizer have 35.9%, 35.96%, and 34.96% for CON, ORG and UNT, respectively, which shows once again the improvement in the chemical quality of the forage in question using green fertilization values close to this NDF result which were found by Pariz et al. (2010). For the ADF, the conventional form presented higher results, with lower value when green fertilization was used about the unplanted plots, demonstrating the ability of this to reduce the cell wall compounds. Results similar to those found by Reis et al. (2013) and Carvalho et al. (2019), who found averages of NDF and ADF values close to those observed in this study.

The LIG content was lower and consequently better for green fertilizer in the organic system with a 3.9% content. Sá et al. (2010) found that 4.5% of lignin for this same species of forage, but cut at 54 days, therefore, low values of lignin may be related to the fact that a good part of the lignin fraction is potentially soluble in the sulfuric acid solution which is may have underestimated the content of this component in the samples evaluated (Migita and Kawamura 1994). However, this result can also be justified by the possible improvement that the green fertilizer provoked in the soil, favoring the chemical quality of the forage.

On the other hand, an increase in crude protein levels and a reduction in forage fiber were also observed by Fagundes et al. (2012a), using *Panicum maximum* cv Tanzania under organic management and by Fagundes et al. (2012b) evaluating organic weeds of Pennisetum purpureum cv. Napier intercropped with *Macroptilium atropurpureum*, D.C., cv Siratro for dairy herd food supplementation. This result corroborates that regardless of the gender used, adequate nutritional maintenance occurs by feeding with forage of the animals in the organic production system.

The MS productivity data found are within the mean of the *Urochloa* and *Stylosanthes* standards as described by several authors each in its experimental specificities and obeying distinct periods over the years.

Urochloa and *Stylosanthes* consortium percentages were distinct between the fertilization forms and in green fertilization. However, for *Urochloa* the percentage with green fertilization, there was not different between the fertilization forms from the one observed without green fertilization, in which the conventional one presented higher establishment value in the plots. This fact is likely to be associated with a possible non-alteration of the physical and biological characteristics of the soil that could have improved the efficiency of the use of the natural rock fertilizers applied in the system of organic fertilization. Also, the lowest percentage rate of legume in intercropping can be attributed to competition for water, light, and nutrients, common among plants of different species.

The *Urochloa brizantha* cv. Marandu, being a plant with greater photosynthetic efficiency (C4 cycle) under tropical conditions and better adapted to low fertility soil conditions, was more competitive than the legume (C3 cycle) (Aroeira et al. 2005; de Andrade et al. 2015). As observed, the percentage of legume in the pasture was always higher under organic management. The average of the 3 years with or without green fertilizer presented values of 12% and 13%, respectively, about the conventional management (11% and 9%), which was even 7% lower than the control in the plots without green fertilizer.

The use of dry matter and nitrogen provided by the use of *Crotalaria juncea*, incorporated to the soil before planting of the pasture, as green fertilizer, which was around 705 kg.ha^{-1} and 24 g/kg, respectively, may have also influenced the productivity of dry matter over the years. The highest percentage of forage legumes in the pasture shows, in general, their ability to grow and compete in soils with lower nutrient availability (Soares et al. 2018). In this sense, this technology is shown to be strategic for use in organic animal production.

The dry matter production of *Stylosanthes* spp. did not change with green fertilization with or without Guandu bean; this same behavior is repeated for plots without green fertilizer; and this shows that regardless of the presence of Guandu bean, the productive behavior of *Stylosanthes* spp. was the same. But the same does not occur when compared with or without green fertilizer about Guandu bean, the graph shows that the lack of Guandu bean (untreatment) promoted the reduction of the production of *Stylosanthes* spp. in areas with green fertilizer and without green fertilizer.

There is a possibility of the Guandu bean in contribution with *Stylosanthes* spp.; in the process of biological nitrogen fixation, this promotes a better contribution of nitrogen molecules, which reflected positively on the growth and dry matter production, a fact that did not occur when the pigeon was not present. According to Barcelos et al. (2008), the release of the biologically fixed nitrogen will respond in large part to the maintenance of grass productivity. The interaction observed between green fertilization and mycorrhizal fungi for dry matter production of *Stylosanthes* spp. can be explained because in areas with green fertilization, the means behave equally independently if there is or not mycorrhiza; this same behavior is repeated for plots without green fertilizer; and this shows that regardless of the presence of mycorrhizas, the productive behavior of *Stylosanthes* spp. with or without green fertilizer was the same.

The same does not occur when comparing the green fertilizer about the presence of mycorrhiza, (untreatment) promoted the reduction of 34.63% of the *Stylosanthes* spp. production with green fertilizer about the plots without green fertilizer. This behavior is due to a possible small microbial population in the soil, which reduced the use of the material used in green fertilization as a source of nutrients.

The green fertilizer versus fertilizer showed values of DM content of 47.50% and 48.63% in the green fertilizer for the conventional and control fertilizer systems, respectively. The organic system present lower DM content (35.21%), that is, did not respond positively to green fertilization.

Analyzing the in vitro digestibility of *Urochloa* dry matter (Table 4) about green fertilizer and fertilization forms, the UNT presented a higher result with green fertilizing (55.91%), but the same did not occur from the ORG system, reflecting the ADF content in that the same treatments presented the lowest values for the green fertilizer. In the treatment without green fertilization, higher values of digestibility with the forms of conventional (62.95%) and organic (62.60%) fertilization were found in contrast, the control presented (56.3%) values similar to those found by Velásquez et al. (2010) and Moreira et al. (2013) which shows that the green fertilization did not favor the digestibility of the grass probably because the fibrous constituents (NDF, ADF, and lignin) are negatively correlated to the digestibility of forages (Brito et al. 2003).

The application or not of mycorrhizal fungi and the presence or absence of Guandu about the DM content demonstrate that as plots with rows of Guandu beans suffer greater DM when inoculated with mycorrhizal fungi about plots without Guandu, even with inoculation. In this study, it can be concluded that the legume may improve the efficiency of inoculated mycorrhizal fungi by up to 11.72% in the DM content of *Urochloa brizantha* cv. Marandu; however, there was no effect between the presence and absence of Guandu bean without inoculation of mycorrhizal fungi, and it is evident that the efficiency of the use of mycorrhizal fungi inoculation for this variable (DM) only occurs when there is the presence of leguminous lines, in this case, the Guandu bean.

The hypothesis that the presence of green fertilizer may have a negative influence on mycorrhizal activity was observed when evaluating the interaction between systems with or without green fertilizer about the presence of mycorrhiza since it was observed that the presence of green fertilizer promoted a 5.19% increase in CP concentration of *Urochloa brizantha* cv. Marandu.

In summary, it can be inferred that the increase in dry mass yield and crude protein contents of forage are important factors to improve the supply and nutritional quality of forage, constituting the technologies used in this work as viable for animal feed in an organic production system.

Conclusions

First, the conventional and organic fertilization forms presented the same behavior for the system conditions with or without green fertilization, indicating that the substitution of chemical inputs by alternative chemical inputs is feasible.

Second, the inoculation of bacteria and mycorrhiza promotes a better production of DM independent of the fertilization system, as well as the presence of the Guandu bean. *Urochloa brizantha* cv. Marandu was superior in chemical bromatological quality with green fertilization in the form of organic fertilization, demonstrating better parameters of protein fractionation. The *Stylosanthes* spp. demonstrated to have better chemical bromatological quality with green fertilization in the system of organic fertilization, the results of fractionation of carbohydrates and nitrogen compounds affirm the superior quality for this species of legume.

Finally, it can be said that, about productivity aspects per hectare and nutritional aspects of forage, organic treatment was as good as conventional treatment. Organic cultivation also showed greater stability in the release of compounds in the soil, indicating that the use of green fertilization, intercropping with legumes, inoculation with mycorrhizae in forage, and fertilization with natural inputs are capable of producing organic forage for animal production. Therefore, it is recommended to use organic production systems intercropped with leguminous.

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