












Tamani grass-legume intercropping can improve productivity and composition of fodder destined to haylage or hay

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ABSTRACT: *This research evaluated the biomass productivity and nutritional value of the haylage and hay from intercropping between Tamani grass and different legume species. For the productive characteristics of the different intercrops, we adopted a randomized block design, for evaluation of the combination of intercropping and conservation technic we used 5 x 2 factorial scheme (five intercrops and two types of conservation techniques). The treatments were Tamani grass as monoculture, and the intercrops of Tamani grass with crotalaria, soybean, cowpea, or pigeon pea. The conservation techniques were haylage (520 g/kg of DM) and hay (870 g/kg of DM). Plants were sown in alternate rows, with 45 cm of spacing between the rows. The parameters evaluated were grass and legume biomass production, canopy height, and haylage and hay chemical composition, and in vitro dry matter digestibility (ivDMD). There were no differences in the total biomass production between the intercrops and TA grass monoculture. The treatments intercropped with cowpea and soybean had the highest legume participation in the mixture, promoting an increase in crude protein and ivDMD content of haylage and hay. Haylage and hay had the same chemical composition, although haylage had higher ivDMD than hay. We concluded that intercropping Tamani grass with soybeans or cowpea maintained total biomass productivity and improved the nutritional value of haylage and hay.*

Key words: *Cajanus cajan, Crotalaria ochroleuca, Glycine max, Panicum maximum, Vigna unguiculata.*

Consórcio da gramínea Tamani com leguminosas pode melhorar produtividade e composição da forragem destinada a silagem ou feno

RESUMO: *Esta pesquisa foi realizada com o objetivo de se avaliar a produtividade da biomassa e valor nutricional da silagem e do feno provenientes do consórcio entre o capim-Tamani e diferentes espécies de leguminosas. Para as características produtivas das diferentes culturas consorciadas, adotamos o delineamento de blocos ao acaso, para avaliação da interação dos consórcios com técnicas de conservação usamos esquema fatorial 5 x 2 (cinco consórcios e duas técnicas de conservação). Os tratamentos foram o capim-Tamani em monocultura, e o consórcio de capim-Tamani associado a crotalária, soja, feijão-caupi ou feijão-guandu. As plantas foram semeadas em fileiras alternadas, com espaçamento de 45 cm. As técnicas de conservação foram silagem pré-secada (520 g / kg de MS) e feno (870 g / kg de MS). Avaliamos a produção de biomassa de gramínea e leguminosa, a altura do dossel, a composição química e a digestibilidade in vitro da matéria seca (ivDMD). Não houve diferenças na produção total de biomassa entre os consórcios e o monocultivo de capim-Tamani. Os tratamentos consorciados com feijão-caupi e soja tiveram maior participação de leguminosas na mistura, promovendo aumento no teor de proteína bruta e ivDMD da silagem pré-secada e feno. A silagem pré-secada e o feno tinham a mesma composição química, embora a silagem pré-secada apresentou maior ivDMD do que o feno. Concluímos que o consórcio de capim-Tamani com soja ou feijão-caupi manteve a produtividade da biomassa total e melhorou o valor nutricional da silagem pré-secada e do feno.*

Palavras-chave: *Cajanus cajan, Crotalaria ochroleuca, Glycine max, Panicum maximum, Vigna unguiculata.*

INTRODUCTION

The use of forage legumes can be a greater protein source in ruminant diet (LIGOSKI et al., 2020). However, when legumes are fed as the sole fodder, as pasture systems, their protein is often used inefficiently (VAN SOEST, 1994). To obtain the best use of legumes protein, it is recommended

to combine them with concentrated supplementation (VAN SOEST, 1994). Thus, conservation techniques for these forages can contribute to their use in feedlot animal diets. Moreover, legumes are often challenging to ensile because of their low sugar content and high buffering capacity (BORREANI et al., 2018). When legumes are used for hay production, it can have a significant leaf loss during

dehydration and a consequent decrease in nutritional quality (HARRIS et al., 2017).

Studies have shown benefits of intercropping grasses (cereals or grasses) and legumes to minimize losses during storage, increase fodder productivity, and improve feed quality for ruminants. According to ADESOGAN et al. (2004), grass-legume intercropping improves N utilization efficiency, by combining cereal N absorption capacity with the legume biological N-fixation capacity, resulting in higher productivity. However, competition between species can lead to an unbalance in the proportion of grass and legume in the canopy, or even to a decrease in biomass productivity if the wrong choices are made concerning species, spatial arrangements, and sowing time (NERES et al., 2011). Regarding forage conservation, FOSTER et al. (2011) concluded that due to the high pH and butyrate concentrations and low lactate and VFA concentrations of cowpea and pigeon pea haylage, indicating the necessity to optimize the fermentation of these forages.

Therefore, we evaluated the biomass productivity, fermentation of haylage, and nutritional value of the haylages and hay from intercropping between Tamani grass and different legume (crotalaria, soybean, pigeon pea, and cowpea). We hypothesized that the intercropping with Tamani grass and forage legumes favors biomass production and improves the fermentation of haylage and nutritional quality of haylages and hay.

MATERIALS AND METHODS

We carried out the field experiment at Embrapa Agropecuária Oeste and analyzed the chemical composition of haylages and hays at the Agricultural Waste Utilization Laboratory at the Federal University of Grande Dourados (UFGD). Both research centers are located in Dourados, MS, Brazil (22 ° 11'55 " S, 54 ° 56'7 " W and 452 m above sea level). The local climate is Cwa (humid mesothermal, with rainy summer), according to the Köppen classification. The temperature, relative humidity, rainfall, and solar radiation observed during the forage growth are in table 1.

The experimental area soil is classified as dystrophic dark red latosol, with a very clayey texture (SANTOS et al., 2018), which presented the following characteristics: Sand: 82.45 g/kg; Silt: 191.33 g/kg; Clay: 720.94 g/kg; pH: 5.12; pH CaCl₂: 4.34; Al₃⁺: 0.68 cmolc/dm³; Ca₂⁺: 2.61 cmolc/dm³; Mg₂⁺: 0.74 cmolc/dm³; H⁺ + Al₃⁺ = 6.31 cmolc/dm³; K⁺ = 0.72 cmolc/dm³; P (Mehlich) = 10.11 mg/dm³; Sum of

bases = 4.07 cmolc/dm³; Cation Exchange Capacity = 10.38 cmolc/dm³; Base Saturation = 39.21%; Total C = 2.52% and Organic Matter = 26.53 g/kg. Based on soil chemical analysis, we applied 2,300 kg/ha of dolomitic limestone (RNV 80%) to raise the soil base saturation to 50%. We applied 150 kg/ha of 04-18-18 (N-P-K) formula as sowing fertilization.

Productive characteristics

The experiment was designed in randomized blocks (based on area declivity and shading) for productive characteristics of the different intercrops. The tested treatments were Tamani grass (TA) monoculture, Tamani grass + *Crotalaria ochroleuca* (TA + Co), Tamani grass + cowpea (TA + Cp) and Tamani grass + Soya (TA + So) and Tamani grass + Pigeon pea (TA + Pp). There were five replications per treatment, totaling 25 experimental plots.

Legume cultivars and Tamani grass were sown on november 19, 2019, using a planter (SHM 1517, Semeato), with 45 cm spacing between plant rows. The grass-legume planting ratio was 1:1, with a row of grass alternated with a row of legumes. Each experimental plot had 14 rows with 10 m long, where we excluded 1.0 m of border defining the sampling area. On the day of the cut (January 22, 2020), we collected the fodder contained in 1.5 linear meters of central rows (grass and legume) at five random points per plot. The collected material was weighed (fresh mass) and sent to the laboratory for subsequent botanical separation (grass and legume). We determined the dry matter (DM) by drying the material in a forced circulation oven at 55 °C for 72 hours. With the DM values and their respective green mass productivities by area, it was possible to calculate the grass and legume total biomass yields.

Conservation techniques

We adopted a complete randomized design to evaluate the conservation technic effect in a 5 x 2 factorial scheme, with five types of intercropping (TA, TA+Co, TA+So, TA+Cp, and TA+Pp), and two types of conservation techniques (haylage and hay).

After plots were sampled, we harvested (5 cm above ground level) the remaining fodder of each plot for haylage and hay production. Plants were harvested at 09:00 am on a sunny day with an average air temperature and relative humidity of 30 °C and 65%, respectively. The fodder from each plot was transported to a greenhouse where manual overturning was performed to achieve homogeneous dehydration rates and to protect the fodder in case

Table 1 - Air temperature, minimum (Min), medium (Med), and maximum (Max), relative air humidity (RH), rainfall, and radiation, observed during the forage growth.

Item	-----Month/Year-----				
		Nov/2019	Dec/2019	Jan/2020	Feb/2020
Temperature, °C	Max	34.00	31.41	32.08	32.34
	Med	27.05	25.24	26.25	26.15
	Min	21.52	20.57	22.15	21.55
RH, %		63.53	76.00	77.45	74.55
Rainfall, mm		108.60	216.80	172.40	147.20
Radiation, MJ/m ² /d		13.08	13.28	13.29	13.20

of raining. After 4 hours of dehydration, fodders reached values between 400 and 500 g/kg of DM. We divided each plot material into two portions. One portion was sent to haylage production and the other one remained for dehydration until 850 g/kg DM for hay production.

We chopped (particle size 2cm) the material for haylage production to facilitate filling and compacting processes. After chopping, the fodders from each plot were homogenized and ensiled. We sampled each treatment at the time of ensiling, approximately 300 g, to determine the chemical composition and *in vitro* DM digestibility (*iv*DMD) and we froze approximately 70 g for further processing and determination of pH. We stored the fodder mass in silos built of PVC tubes measuring 10 cm in diameter and 50 cm in height, with a volume of 3.8 L. The haylage was compacted at an average density of 520 kg/m³ manually with wooden sticks. After filling, we sealed the silos with double-sided plastic canvas (black and white) and adhesive tape and stored it in the laboratory at room temperature for 90 days. For the calculation of fermentative losses, all experimental silo components and ensiled fodder mass were weighed before and after ensiling. We calculated the gas losses (g/kg of ensiled DM) and DM recovery (g/kg of ensiled DM) according to the equations of LI et al. (2017).

After opening, haylage was removed and homogenized for sample collection. A sample (300 g) was dried in an oven with forced circulation at 55 °C for 72 h DM calculations. Then, it was ground in a Willey mill with a 1.0 mm mesh sieve to determine haylage chemical composition. We determined the contents of DM, ash (method 942.05), and crude

protein (CP, method 976.06) according to AOAC (2005). We analyzed the contents of neutral detergent fiber assayed with a heat stable amylase (aNDF), acid detergent fiber (ADF), hemicellulose, cellulose, and lignin according to the protocols suggested by MERTENS, (2002). We determined the *iv*DMD according to the methodology described by TILLEY and TERRY, (1963).

Another haylage sample (70 g) was collected and frozen to determine pH and organic acid profile. Samples (pre-dry fodder and haylage) were defrosted, diluted (25 g fodder in 225 mL distilled water), and homogenized in an industrial blender for approximately 5 minutes. The extract was used to determine pH using a digital potentiometer (mPA210 MS Tecnoyon). We filtered a portion of the extract through a paper filter, centrifuged it for 15 min at 10,000 rpm, and frozen the supernatant at -20 °C to further organic acids analysis. Organic acids were determined by gas chromatography with a mass detector (GCMS QP 2010 Plus, Shimadzu, Kyoto, Japão) and a capillary column (Stabilwax, Restek, Bellefonte, EUA, 60 m, 0,25 mm Ø, 0,25 µm de polietileno cross bond carbowax glicol). We determined lactic acid content by the colorimetric method proposed by PRYCE (1969). The content of ammoniacal nitrogen (N-NH₃) was determined according to the methodology described by CARLSON (1978).

The portion of fodder remained in the greenhouse, was kept approximately 36 hours to until reaching values between 850 to 900 g/kg of DM. In sequence, the material was collected, baled, identified, and stored in a covered and airy shed. Five bales were produced (one for each plot) with

approximately 10 kg per treatment. After 90 days of storage, we collected samples (approximately 100g) of each bale to determine the contents of DM, ash, CP, aNDF, ADF, hemicellulose, cellulose, lignin, and *iv*DMD, following the methodologies previously described.

Data evaluation

The data were analyzed using the RStudio statistical program (R, 2019) through the PROC MIXED procedure. Means of intercrop productive characteristics were compared using the Scott Knott test, with a 5% significance level, according to the following model: $Y_{ki} = \mu + \beta_k + S_i + \varepsilon_{ik}$, where: Y_{ki} = dependent variable, μ = average mean, β_k = block effect (random effect; $k = 1, 2, 3, 4$ e 5), S_i = intercropping effect (fixed effect of TA, TA+Co, TA+So, TA+Cp, and TA+Pp), and ε_{ik} = random error associated with each observation.

To evaluate the intercropping effects on the fodder conservation technic (haylage and hay), means were compared using the Scott Knott test, with a significance level of 5%. The means of fodder conservation techniques were compared using the F test, with a significance level of 1%, according to the following model: $Y_{ijk} = \mu + I_i + C_j + I * C_{ij} + \varepsilon_{ijk}$, where: Y_{ijk} = dependent variable, μ = average mean, I_i = intercropping effect (fixed effect of TA, TA+Co, TA+So, TA+Cp, and TA+Pp), C_j = fodder conservation technic effect (fixed effect of haylage and hay), $I * C_{ij}$ = effect of the interaction between intercropping and fodder conservation

technic, and ε_{ijk} = random error associated with each observation. Pearson correlation was performed between fermentation parameters with nutritional value, composition, and *in vitro* digestibility, and it was presented as supplementary results.

RESULTS AND DISCUSSION

The temperature was higher throughout the forage development period than the region historical values (FIETZ and FISCH, 2008); the maximum temperature was higher for november and february, while the minimum and average temperatures were higher in november, january, and february. In addition, the relative air humidity was lower than the historical values (73-84 %) in the study period. However, the rainfall was greater in december, january, and february (FIETZ and FISCH, 2008).

The highest grass production was observed in TA monoculture ($P < 0.01$), followed by TA+Pp and TA+Co, without differences between them (Table 2). Intercrop productive efficiency is affected by several environmental and agronomic factors, which can significantly interfere with competition between plants in the canopy (LIGOSKI et al., 2020). Pigeon pea was traditionally grown in a mixture of cereals but developed to have higher-yielding and early maturing, resulting in medium plant height and larger leaf area (SINGH et al., 2021). Moreover, *C. ochroleuca* has an erect growth that varies from 1.5 to 2 meters in height, with an expressive proportion of stem in the biomass composition and narrow

Table 2 - Grass and legume total and specific productivity, ratio, and plant heights from the intercropping of Tamani grass and different legumes.

Item	TA	TA+Co	TA+Cp	TA+So	TA+Pp	SEM	P-value
Total production, kg/ha	5,721	4,987	5,713	5,497	5,431	119.5	0.08
TA production, kg/ha	5,721 ^a	3,029 ^b	2,102 ^d	2,759 ^c	3,359 ^b	264.5	<0.01
Legume production, kg/ha	-	1,957 ^c	3,610 ^a	2,737 ^b	2,072 ^c	251.1	<0.01
TA/Legume ration	-	1.55 ^a	0.58 ^c	1.01 ^b	1.62 ^a	0.180	<0.01
Grass heigh, m	0.99 ^b	1.06 ^a	0.95 ^b	0.97 ^b	1.10 ^a	0.010	<0.01
Legume heigh, m	-	1.46 ^a	0.81 ^b	0.88 ^b	1.35 ^a	0.110	<0.01

TA = Tamani grass monoculture; TA+Co = Tamani grass + *Crotalaria ochroleuca*; TA+Cp = Tamani grass + Cowpea; TA+So = Tamani grass + Soybean; TA+Pp = Tamani grass + Pigeon pea; SEM = standard error of mean. Means followed by different letters differ from each other by the Scott Knott test at 5% probability.

leaves (AMABILE et al., 2000). The adaptation of pigeon pea to intercropping and the thin leaves of *C. ochroleuca* may have favored Tamani grass growth.

The treatments TA+Cp and TA+So had the lowest grass yields, with the lowest value for TA+Cp. Conversely, legume production had the opposite yield, with the highest productivity observed for TA+Cp ($P < 0.01$), followed by TA+So, and lastly by TA+Co and TA+Pp. According to NAMATSHEVE et al. (2020), intercropping with cowpea generally decreased cereal crop yields at different magnitudes. In addition, soya in intercropping systems showed high phenotypic plasticity to complement light capture (ZHOU et al. 2019). These characteristics of these crops could explain the highest productivity of the legume in these intercropping.

Despite variations in individual biomass production between TA grass and legumes, there were no differences ($P = 0.08$) in the total biomass production ($5,470 \pm 597$ kg/ha) of intercrops. The similarity fodder biomass production demonstrates a competition between species present in the intercropping. To increase fodder biomass production in the intercropping, the main crop must have a large size and higher growth speeds than the secondary crop; thus, the main crop will dominate the area and increase its productivity (ESKANDARI & GHANBARI, 2009). Furthermore, it is recommended that secondary crops be sown between the main crop rows, minimizing competition for space and allowing satisfactory growth (AKHTAR et al., 2013).

Legume and grass heights were higher ($P < 0.01$) for TA+Pp intercrop. Pigeon pea was developed to have a higher relative growth rate (SINGH et al., 2021). The most recommended species for hay and haylages are short grasses, which often have growth rates very close to forage legumes (SAIA et al., 2016). NERES et al. (2011), evaluating canopies of Tifton 85 intercropped with oat and ryegrass, observed a decrease in biomass production compared to the monoculture (Tifton 85). According to the authors, the low yields occurred due to the season that they conducted the experiment (winter-spring) and the low shading tolerance of Tifton 85. These results demonstrated the importance of correctly choosing the species that will compose the intercrop and the sowing season, always aiming to balance fodder biomass production nutritional value.

The different biomass production between TA grass and legume changed the TA/Legume ratio ($P < 0.01$). We observed the lowest TA/Legume ratio (0.58) in TA+Cp with 63% legume participation in the final biomass composition. These results

indicated grass dominance in most intercrops. However, cowpea had the highest participation in the intercropped species, showing the most competitive among all evaluated legumes. It is worth mentioning that soybean also had high participation in biomass production (49.9%). According to VASCONCELOS et al. (2020), TA grass has small and narrow leaf blades, making it uncompetitive in systems intercropped with legumes. Conversely, MACHADO et al. (2017) did not observe a reduction in soybean grain productivity when TA grass was sown between the soybean rows, with a sowing delay of 21 days with soybean emergence. Thus, we can consider TA grass associated with cowpea or soybean as good intercrop options, mainly when it intends to increase the legume participation in the biomass.

There were no differences ($P > 0.05$) between intercrops in DM recovery, gas losses, and initial pH for haylage (Table 3). The mean values of DM recovery from the haylages were higher than 970 ± 19 g/kg of the ensiled DM, a value consistent with those reported in the literature (CUNHA et al., 2020). According to BORREANI et al. (2018), pre-drying reduces the fodder water activity, inhibiting secondary fermentation, responsible for the highest DM losses in tropical grass and legume silages. Pre-drying also avoids effluent loss, which is very common in tropical grass silages, liable for soluble nutrient losses (ORRICO JUNIOR et al., 2020). The values referring to gas losses were below 20.2 ± 5.5 g/kg, supporting the high DM recovery values observed. We also observed a negative correlation (Table 4) between gas losses and the concentration of lactic and acetic acid (-0.41 , $P = 0.05$ and -0.43 , $P = 0.03$, respectively), indicating better silage conservation (less gas loss) when these acids are in higher concentration.

For the final pH, we observed higher values ($P < 0.01$) for haylages from TA+So and TA+Pp (5.13 and 5.10, respectively). According to MUCK et al. (2010), pH values equal to or less than 4.0 are necessary to guarantee the process stability and the ensiled fodder quality. However, haylages have lower production of organic acids due to the insufficient development of microorganisms, resulting in higher pH values (MÜLLER et al., 2014). In a review on the main chemical parameters of silages, KUNG et al. (2018) concluded that, for haylages with DM contents between 450-550 g/kg of DM, it is expected average pH values between 4.7 and 5.0. These values were close to those obtained in the present experiment, with pH ranging from 4.53 to 5.13. Although, pH is an indicator of fermentation process quality, this parameter was not correlated with the lactic and

Table 3 - Fermentation parameters and chemical composition of haylages from Tamani grass intercropping with different legumes.

Item	TA	TA+Co	TA+Cp	TA+So	TA+Pp	SEM	P-value
DM, g/kg of fresh	499.3 ^b	528.1 ^a	455.1 ^b	558.0 ^a	573.7 ^a	11.78	<0.01
DM recover, g/kg DM	982.2	975.5	984.7	960.2	968.9	4.01	0.13
Gas loss, g/kg initial DM	20.2	17.5	14.5	15.4	20.2	1.15	0.45
Initial pH	7.63	7.58	6.63	7.65	7.06	0.189	0.85
Final pH	4.53 ^b	4.80 ^b	4.54 ^b	5.13 ^a	5.10 ^a	0.074	<0.01
Lactic acid, g/kg DM	22.4 ^b	18.9 ^b	42.4 ^a	39.1 ^a	19.2 ^b	2.39	<0.01
Acetic acid, g/kg DM	28.9 ^b	32.9 ^b	52.2 ^a	42.4 ^a	40.2 ^a	2.53	<0.01
Lactic:acetic ratio	0.8 ^b	0.6 ^c	0.8 ^b	0.9 ^a	0.4 ^d	0.04	<0.01
Propionic acid, g/kg DM	2.7 ^b	2.9 ^b	4.2 ^b	3.7 ^b	5.9 ^a	0.33	<0.01
Butiric acid, g/kg DM	1.3 ^a	0.9 ^b	0.8 ^b	0.8 ^b	0.9 ^b	0.05	<0.01
Iso-Valeric acid, g/kg DM	4.6	4.1	12.2	9.8	4.7	1.21	0.24
Valeric acid, g/kg DM	8.9	10.0	14.5	12.2	11.6	0.85	0.10
N-NH ₃ , g/kg TN	103.6	71.6	106.1	114.3	137.1	7.23	0.22

TA = Tamani grass monoculture; TA+Co = Tamani grass + *Crotalaria ochroleuca*; TA+Cp = Tamani grass + Cowpea; TA+So = Tamani grass + Soybean; TA+Pp = Tamani grass + Pigeon pea; SEM = standard error of mean; DM= dry matter; N-NH₃= amoniacal nitrogen; TN= total nitrogen. Means followed by different letters differ from each other by the Scott Knott test at 5% probability.

acetic acids ($P > 0.05$), the acids with the highest pKa and in higher concentrations in haylage, since this variable depends on the material buffering capacity (KUNG et al., 2018).

The highest lactic acid content ($P < 0.01$) and lactic: acetic ratio ($P < 0.01$) occurred in the TA+Cp and TA+So. We observed the lowest values of acetic acid content ($P < 0.01$) for TA and TA+Co. In general, legumes have a higher buffering capacity. Thus, the greater legume participation in the ensiled mixture, the higher the organic acid content is necessary to promote pH reduction (JAHANZAD et al., 2016). This explains why TA monoculture haylages presented pH similar to TA+Cp intercrop but with lower production of lactic and acetic acids. BILDIRICI et al. (2009) had similar results; they observed the highest organic acid production for silages with legume proportions above 40 % in the ensiled mixture.

The low lactic acid content and lactic: acetic acid ratios observed in this research indicate an acetic fermentation predominance. This result may be linked to a more significant *Enterobacteria* and *Bacilli* activity in fermentation since the action of these microorganisms is only inhibited when the pH

value is below 4.5 (MUCK, 2010), which did not occur in any of the tested treatments. The TA+Pp had the highest propionic acid content ($P < 0.01$). Conversely, butyric acid content was higher for TA ($P < 0.01$). Also, the butyric acid presented a negative correlation (-0.72 , $P < 0.01$) with the CP, indicating a possible degradation; however, there is no correlation ($P > 0.05$) between butyric acid and N-NH₃. The iso-valeric acid and valeric content did not differ ($P > 0.05$) between intercrops. In general, the haylages assessed in this experiment had N-NH₃ range close to the limit of 100 g/kg of total nitrogen, with no difference ($P > 0.05$) between intercrops, which according to KUNG et al. (2018), indicates low proteolysis. Similar to the FOSTER et al. (2011) study that evaluated the nutritional and fermentative quality of bahia grass or legumes (perennial peanut, annual peanut, cowpea, and pigeon pea) haylages, ensiled as individual crop.

There was no interaction ($P > 0.05$) between intercrops and conservation technic on the chemical composition and *ivDMD* (Table 5). The grass-legume intercropping aims are to increase the fodder CP content (LIGOSKI et al., 2020; NERES et al., 2011; SAIA et al., 2016). For both haylage and

Table 4 - Pearson correlation between fermentation parameters, nutritional value, composition, and *in vitro* digestibility.

	Gas loss	Final pH	DM	CP	NDF	ADF	Cell	Hem	Lig	ivDMD	Lactic acid	Acetic acid	Propionic acid	Butiric acid	Iso-Valeric acid
Final pH	-0.04														
	P=0.84														
DM	0.26	0.51													
	P=0.21	P=0.01													
CP	-0.25	0.29	-0.02												
	P=0.24	P=0.16	P=0.92												
NDF	0.40	-0.14	0.05	-0.83											
	P=0.05	P=0.49	P=0.83	P<0.01											
ADF	0.36	-0.12	0.10	-0.88	0.92										
	P=0.08	P=0.57	P=0.63	P<0.01	P<0.01										
Cell	0.40	-0.14	0.05	-0.83	1.00	0.92									
	P=0.05	P=0.49	P=0.83	P<0.01	P<0.01	P<0.01									
Hem	0.35	-0.26	-0.09	-0.85	0.90	0.88	0.90								
	P=0.09	P=0.22	P=0.67	P<0.01	P<0.01	P<0.01	P<0.01								
Lig	0.25	-0.07	0.14	-0.78	0.68	0.91	0.68	0.71							
	P=0.23	P=0.74	P=0.49	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01							
ivDMD	-0.47	-0.05	-0.28	0.84	-0.78	-0.88	-0.78	-0.72	-0.84						
	P=0.02	P=0.83	P=0.17	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01						
Lactic acid	-0.41	-0.06	-0.33	0.69	-0.72	-0.76	-0.72	-0.62	-0.66	0.76					
	P=0.05	P=0.79	P=0.10	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01					
Acetic acid	-0.43	-0.08	-0.19	0.54	-0.65	-0.53	-0.65	-0.51	-0.32	0.48	0.49				
	P=0.03	P=0.69	P=0.37	P<0.01	P<0.01	P<0.01	P<0.01	P<0.01	P=0.13	P=0.02	P=0.01				
Propionic acid	-0.04	0.29	0.19	0.24	-0.34	-0.10	-0.34	-0.12	0.17	0.05	0.01	0.48			
	P=0.84	P=0.16	P=0.36	P=0.25	P=0.10	P=0.62	P=0.10	P=0.57	P=0.41	P=0.81	P=0.95	P=0.01			
Butiric acid	-0.03	-0.43	-0.09	-0.72	0.44	0.48	0.44	0.49	0.44	-0.47	-0.36	-0.13	-0.16		
	P=0.88	P=0.03	P=0.67	P<0.01	P=0.03	P=0.02	P=0.03	P=0.01	P=0.03	P=0.02	P=0.08	P=0.53	P=0.45		
Iso-Valeric acid	-0.31	-0.15	-0.24	0.38	-0.48	-0.50	-0.48	-0.43	-0.43	0.37	0.55	0.35	-0.15	-0.07	
	P=0.13	P=0.46	P=0.25	P=0.06	P=0.02	P=0.01	P=0.02	P=0.03	P=0.03	P=0.07	P<0.01	P=0.09	P=0.47	P=0.74	
Valeric acid	-0.32	0.04	-0.13	0.43	-0.39	-0.32	-0.39	-0.38	-0.19	0.32	0.42	0.46	0.03	-0.19	0.56
	P=0.12	P=0.85	P=0.52	P=0.03	P=0.05	P=0.11	P=0.05	P=0.06	P=0.36	P=0.12	P=0.04	P=0.02	P=0.90	P=0.36	P<0.01

Gas loss in g/kg of initial DM; Chemical composition, ivDMD, and acids in g/kg of DM; DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; Cell = cellulose; Hem = hemicellulose; Lig = lignin; ivDMD = *in vitro* dry matter digestibility. Variables with non-significative correlations were removed from the data presentation.

Table 5 - Chemical composition and *in vitro* dry matter digestibility of haylage and hay from the intercropping of Tamani grass and different legumes.

Item, g/kg DM	-----Intercropping-----					--Conservation techniques--		SEM	-----P-value-----		
	TA	TA+Co	TA+Cp	TA+So	TA+Pp	Haylage	Hay		I	C	I*C
Ash	99.8	90.0	83.3	87.7	87.0	86.8	92.3	01.44	0.48	0.42	0.17
CP	57.4 ^c	85.2 ^b	107.9 ^a	109.2 ^a	81.4 ^b	88.4	84.1	03.61	<0.01	0.07	0.07
NDF	700.8 ^a	671.0 ^b	608.9 ^c	615.6 ^c	658.8 ^b	660.2	641.8	08.78	<0.01	0.09	0.09
ADF	310.5 ^a	297.8 ^b	284.9 ^c	288.9 ^c	297.7 ^b	295.2	296.7	05.54	<0.01	0.69	0.51
Cell	239.2 ^a	246.0 ^a	214.2 ^c	218.7 ^c	224.2 ^b	238.4	218.5	03.11	<0.01	0.37	0.14
Hem	394.8 ^a	354.1 ^b	306.0 ^c	298.2 ^c	361.1 ^b	340.6	345.1	07.97	<0.01	0.87	0.14
Lig	77.6 ^a	69.6 ^b	54.2 ^c	47.2 ^c	67.8 ^b	56.8	75.4	02.83	<0.01	0.89	0.08
ivDMD	628.7 ^c	641.5 ^b	719.4 ^a	703.0 ^a	618.5 ^c	669.6	652.9	01.21	<0.01	0.03	0.33

TA = Tamani grass monoculture; TA+Co = Tamani grass + *Crotalaria ochroleuca*; TA+Cp = Tamani grass + Cowpea; TA+So = Tamani grass + Soybean; TA+Pp = Tamani grass + Pigeon pea; SEM = standard error of mean; I = intercropping; C = Conservation technic; I*C = Interaction between intercropping and conservation technic; DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; Cell = cellulose; Hem = hemicellulose; Lig = lignin; ivDMD = *in vitro* dry matter digestibility. Means followed by different letters differ from each other by the Scott Knott test at 5% probability.

hay, we observed higher CP content in the intercrops when compared to TA monoculture (Table 3). We observed the highest CP content ($P < 0.01$) in TA+So and TA+Cp and the inverse result for the content of fibrous fractions ($P < 0.01$), with the highest fiber content for TA monoculture, as a result, from the legume participation in the biomass in intercropping. There is a negative correlation between CP content and fibrous fraction (-0.78 to -0.88, $P < 0.01$; Supp. (Table 1). Thus, using these grass-legume mixtures allows greater fodder participation in the diet composition for ruminants and reduces the amount of protein concentrates inclusions to active the animal requirements (ADESOGAN et al., 2004).

The intercrops also resulted in haylages and hays with lower fibrous fractions and higher ivDMD than TA monoculture. There is negative correlation between the ivDMD and fibrous fractions (-0.72 to -0.88, $P < 0.01$; Supp. (Table 1) in the haylage. Among the intercrops, the treatments TA+Co and TA+Pp had the highest fibrous fraction ($P < 0.01$), while the conserved fodder from TA+So and TA+Cp had the lowest values. The opposite behaved was observed for the ivDMD; the TA+So and TA+Cp had ivDMD 14.81% higher than TA+Pp and TA monoculture. According to VAN SOEST (1994), tropical grasses (C4) have "Kranz" type anatomy, which is composed of thick cell wall structures, which increases fiber

concentration and reduces microbial degradation in the rumen when compared with legume (C3) anatomical characteristics. NERES et al. (2012) also observed lower fiber contents and higher ivDMD in mixed hays compared to grass-exclusive hay. We also observed a high correlation (0.75, $P < 0.01$) between ivDMD and lactic acid content. The high lactic acid content probably allows conservation of water-soluble carbohydrates, which results in high ivDMD for the intercropping with cowpea and soya. Similar to that was reported by WEI et al. (2021), the proso Millet had high water-soluble carbohydrates and consequently high ivDMD.

When comparing the chemical composition only between intercropped treatments, it is noticed that TA+Co and TA+Pp were the ones that presented less satisfactory results, mainly concerning CP content and fiber fractions. This result may be associated with the competition effect between species in intercrops, which forced legumes to change the morphogenic and structural characteristics to compete for light, space, and nutrients. Furthermore, the highest grass and legume heights were observed precisely in the TA+Co and TA+Pp, demonstrating that these species had to increase their stem elongation rates to compete for space and solar radiation. Thus, the higher stem proportions observed in these intercrops may be responsible for the lower nutritional value of these

fodders, mainly when it corresponds to the lignified stems of legumes (VAN SOEST, 1994).

The *iv*DMD were influenced by conservation technic ($P < 0.01$). When comparing fodder conservation techniques ($P < 0.05$), haylage had high *iv*DMD, with an increase of 2.55% compared to hay *iv*DMD. Silages generally have higher nutritional value than hays, even when produced under similar cutting conditions (species, age, time of year, and environment). Several reasons explain this superiority: 1) more significant leaf losses that occur during hay rollers reduce CP content and increase hay fiber, especially in legume hays (HARRIS et al., 2017); 2) prolonged periods of dehydration in the field allow greater intrinsic consumption of soluble carbohydrates (respiration of plant cells and aerobic microbial activities) leading to an increase in the fibrous fractions of hays (NERES et al., 2011); 3) prolonged storage can lead to hay moistening (variations in relative humidity) and fungi growth, which lead to significant losses in fodder quality (MÜLLER, 2018); 4) the action of fermentative microorganisms in haylage processes improve passage rate and ruminal degradation when compared to hay, especially regarding fiber utilization (ARROQUY et al., 2014). However, higher *iv*DMD was observed only for the haylages in this research, with no significant differences for the other evaluated parameters. Probably, the fact that the whole haymaking process (harvesting, turning, and baling) was done manually and in a controlled environment (greenhouse), there were fewer losses when compared to mechanically processed hays under field changes concerning climatic conditions (CARTER, 1960). Even so, improvements in *iv*DMD contributed to a better ruminal degradability of the fodder when submitted to fermentation processes, as proposed by ARROQUY et al. (2014).

CONCLUSION

Tamani grass as a monoculture and all grass-legume intercropped tested had similar total biomass production. Grass-legume intercropped resulted in haylages and hays with higher nutritional values. Tamani grass intercropped with soybean or cowpea are promising options for farmers who want to increase crude protein in ruminant diets based on forage sources. However, intercropped with cowpea provides a better fermentation profile than soybean. Based on *in vitro* dry matter digestibility, the haylage is the most proper technic for conserving these intercrops.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare no conflict of interest. The founding sponsors had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, and in the decision to publish the results.

AUTHORS' CONTRIBUTIONS

All authors contributed equally for the conception and writing of the manuscript. All authors critically revised the manuscript and approved of the final version.

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