



Nutritional diversity of *Brachiaria ruziziensis* clones

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ABSTRACT: The aim of this study was to evaluate the nutritional diversity of *Brachiaria ruziziensis* clones through chemical composition and *in vitro* kinetics of ruminal fermentation. Twenty three clones of *Brachiaria ruziziensis* were used (15, 16, 46, 174, 411, 590, 651, 670, 768, 776, 844, 859, 950, 965, 970, 975, 1067, 1093, 1296, 1765, 1806, 1894 and 1972) and *Brachiaria ruziziensis* cv. 'Kennedy', *Brachiaria brizantha* cv. 'Marandu' and *Brachiaria decumbens* cv. 'Basilisk' as controls within 27 days of harvesting. The experimental design used randomized blocks with 26 treatments (genotypes) and three replications. Evaluation of the nutritional divergence was performed using principal components analysis, based on the following discriminatory variables: *in vitro* dry matter digestibility (IVDMD), neutral detergent fiber (NDF), lignin, crude protein (CP), degradation rate of non-fibrous carbohydrates (KdNFC) and degradation rate of fibrous carbohydrates (KdFC). The evaluation of the nutritional diversity of *Brachiaria* genotypes was based on the two main components (IVDMD and NDF), which explains 96.2% of the total variance. Variables of lower contribution to the discrimination of the clones were as degradation rates of the fibrous and non-fibrous carbohydrates. In the agglomerative hierarchical grouping analysis, five distinct groups were identified, where V group, formed by clones 46, 768 and 1067 have higher values of IVDMD compared to the other clones.

Key words: digestibility, discriminating variables, main components, nutritional value.

Divergência nutricional de clones de *Brachiaria ruziziensis*

RESUMO: O objetivo deste estudo foi avaliar a divergência nutricional de clones de *Brachiaria ruziziensis* através da composição química e cinética de fermentação ruminal *in vitro*. Os tratamentos consistiram de 23 clones de *Brachiaria ruziziensis* (15, 16, 46, 174, 411, 590, 651, 670, 768, 776, 844, 859, 950, 965, 970, 975, 1067, 1093, 1296, 1765, 1806, 1894 e 1972), e as testemunhas *Brachiaria ruziziensis* cv. 'Kennedy', *Brachiaria brizantha* cv. 'Marandu' e a *Brachiaria decumbens* cv. 'Basilisk', colhidas com 27 dias de rebrota. O delineamento experimental utilizado foi o de blocos casualizados, com 26 tratamentos (genótipos) e três repetições. A avaliação da divergência nutricional foi realizada utilizando-se a análise de componentes principais e agrupamento aglomerativo hierárquico. Com base nas seguintes variáveis discriminatórias: digestibilidade *in vitro* da matéria seca; fibra em detergente neutro; lignina; proteína bruta; taxa de degradação de carboidratos não fibrosos e; taxa de degradação de carboidratos fibrosos. A avaliação da divergência nutricional dos clones de *B. ruziziensis* baseou-se nos dois primeiros componentes principais (DIVMS e FDN), explicando 96.2% da variância total. As variáveis de menor contribuição para a discriminação dos clones foram as taxas de degradação dos carboidratos fibrosos e não fibrosos. Na análise de agrupamento aglomerativo hierárquico foram identificados cinco grupos distintos, em que o grupo V, formado pelos clones 46, 768 e 1067 destacou-se em relação aos demais por apresentar valores superiores de digestibilidade *in vitro* da matéria seca.

Palavras-chave: componentes principais, digestibilidade, valor nutricional, variáveis discriminatórias.

INTRODUCTION

In Brazil, beef and dairy cattle production is based primarily on grass-feeding systems (around 90%), making pasture grasses the main source of animal feed. With growth in the livestock sector, the search for foods that combine high production and high-quality has been increasing. However, only a few varieties meet the requirements, demonstrating the importance of the introduction of genetically improved cultivars.

Although, the number of forage species available in Brazil is high, *Brachiaria* and *Panicum* occupy the largest area. Among the *Brachiaria* species cultivated in Brazil, *Brachiaria ruziziensis* is not widely used in the country despite showing promise for breeding programs with high nutritional quality, good adaptation in crop-livestock-forest integration system (CLFIS), suitable ground cover with direct planting, and as the only diploid sexual species, which allows variability between generations for selection

of superior genotypes. However, this forage species also has some unfavorable characteristics, such as susceptibility to be attacked by spittlebug, with low productivity, and reduced adaptation to less fertile and acidic soils (DIAS et al., 2013).

Multivariate analysis has been used to evaluate nutritional diversity in forage species (AZEVEDO et al., 2003; FREITAS et al., 2006), helping to identify genotypes with genetic differences that produce progeny with greater heterogeneity, thus increasing the likelihood of obtaining superior individuals in segregating generations (CRUZ et al., 2012; SHIMOYA et al., 2002).

Selecting for good performance and high nutritional value improves efficiency of a breeding program (CRUZ et al., 2012). In this context, the objective was to evaluate the nutritional diversity of 23 *Brachiaria ruziziensis* clones in the breeding program at EMBRAPA.

MATERIALS AND METHODS

The experiment was performed at the Experimental Complex Multiuser of Bioefficacy and Sustainability of Livestock and at the experimental farm of José Henrique Bruschi of EMBRAPA Dairy Cattle in Coronel Pacheco-MG, Brazil (23°35'16" S, 43°15'56" W, altitude of 426m). The climate corresponds to the Cwa type (mesothermal) in the Koppen classification and the soil of the experimental area is classified as Red-Yellow Alic Argisol (SANTOS et al., 2006).

The experimental design used randomized blocks with 26 treatments (genotypes) and three replications. Treatments consisted of 23 clones of *Brachiaria ruziziensis*, from the forage breeding program of EMBRAPA, represented by IDs: 15, 16, 46, 174, 411, 590, 651, 670, 768, 776, 844, 859, 950, 965, 970, 975, 1067, 1093, 1296, 1765, 1806, 1894 and 1972. *Brachiaria ruziziensis* cv. 'Kennedy', *Brachiaria brizantha* cv. 'Marandu' and *Brachiaria decumbens* cv. 'Basilisk' were used as a control.

Plants in each plot were cut within 27 days of growth, at an average height of 10cm with the aid of motorized costal mower. After collection, samples were weighed fresh and drying ovens with forced air circulation at 65°C for 72 hours. They were ground in a Wiley mill with 1 mm sieve and stored in polyethylene bottles for later composition analyses.

The contents of dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), lignin, and *in vitro* dry matter digestibility (IVDMD), were determined by

near infra-red spectroscopy (NIRS) in the ruminant nutrition laboratory at the Animal Science section of EMBRAPA Dairy Cattle, Juiz de Fora, MG, Brazil.

The *in vitro* rumen fermentation kinetics was performed by the semiautomatic gas production technique following the procedures described in MAURICIO et al. (1999). For this evaluation, 50mL glass flasks were utilized and 0.32g of the substrate to be tested were incubated in filter bags F57 (Ankon®). Twenty-eight mL of buffered culture medium and rumen fluid, prepared the previous day (MENKE & SEINGASS, 1987) and kept pressurized under CO₂, were added. Flasks were sealed with a silicone stopper to avoid contamination and fermentation and refrigerated at 4°C. Five hours before inoculation with rumen fluid, the bottles were placed in a room at 39°C.

The rumen inoculate was collected from three fistulated Holstein x Gyr dry cows with 500±15kg average body weight. Diet comprised of pasture (*Brachiaria decumbens*) supplemented with 15kg/day of corn silage and 2.0kg of concentrate a day. Rumen fluid was collected in the morning before feeding through the ruminal cannula and then filtered through double layers of cheesecloth and maintained at 39°C. Inoculum of three cows was pooled.

Three milliliters of rumen fluid was added to the flasks containing samples and buffered culture medium. Finally, flasks were sealed with a silicone stopper and aluminum washers to avoid gases escaping. Triplicates of each sample were incubated and kept heated at 39°C room. Flasks containing only inoculum and culture medium were used as a blank.

Pressure readings were taken using a pressure transducer (DPI 705 – GE) at 2, 4, 6, 8, 10, 12, 14, 17, 20, 24, 28, 34, 48, 72 and 96 hours after inoculation. The PSI values were converted to volume according to the equation: Volume = -0.0125x²+3.6015x - 0.1118; R²=0.9874, established from the laboratory conditions. Production volumes were adjusted to g of substrate (based on DM) incubated and the values obtained were corrected for blanks (flasks without substrate at each incubation time).

A mathematical description of rumen fermentation kinetics was estimated using *in vitro* gas production. The bi-compartmental model was used and fitted to the curve of cumulative gas production (SCHOFIELD et al., 1994) as described below:

$$V(t) = \frac{VFNFC}{(1 + \exp(2 \cdot 4 \cdot kdNFC \cdot (T-L)))} + \frac{VFFC}{(1 + \exp(2 \cdot 4 \cdot kdFC \cdot (T-L)))}$$

Where: V (t) = total gas accumulated at time t; VFNFC is equivalent to the maximum

volume of gases from the NFC fraction (mL); VFFC is the maximum volume of the gases from the FC fraction (mL); kdNFC is the degradation rate (% h) of NFC; kdFC is the degradation rate (% h) of FC; and T and L are the incubation (hours) and lag (hours) times, respectively.

The data of bromatological composition were submitted to the univariate analysis through the Minitab 16 program and the averages compared by the Tukey test at 5% of probability. Principal components analyses and agglomerative hierarchical clustering (complete linkage) were conducted in Minitab 16 to evaluate the nutritional divergence between genotypes. Euclidean distance using standardized mean was used as a basic measure of similarity.

RESULTS AND DISCUSSION

The dry matter (DM), *in vitro* dry matter digestibility (IVDMD), and crude protein (CP) variables had a significant effect among the evaluated clones ($P < 0.05$; Table 1). The DM content ranged from 202.6 to 147.0g/kg DM, with the highest DM content observed for clone 859, with a mean of 202.6g/kg DM, whereas the clone presented lower IVDMD, with a mean of 577.5g/kg DM ($P < 0.05$). The highest values of IVDMD were observed for clones 16, 46, 768, 970 and 1067, with a mean of 679.7 to 692.1g/kg DM ($P < 0.05$). Clones showed high protein content (range 164.9 to 130.4g/kg DM) and clone 15 had the highest value of protein.

Table 1 - Chemical compositions (g/kg DM) of the 23 clones and *Brachiaria ruziziensis* cv. 'Kennedy', *Brachiaria brizantha* cv. 'Marandu' and *Brachiaria decumbens* cv. 'Basilisk'.

Clones	DM (g/kg)	SD	IVDMD (g/kg)	SD	NDF (g/kg)	SD	ADF (g/kg)	SD	Lignin (g/kg)	SD	CP (g/kg)	SD
15	162.4 _{abc}	7.1	661.5 _{ab}	7.0	602.8 _a	13.3	306.9 _a	10.3	44.7 _a	4.1	164.9 _a	19.2
16	160.8 _{abc}	15.0	692.1 _a	29.4	580.4 _a	6.9	288.5 _a	4.1	43.0 _a	1.0	173.5 _{ab}	18.0
46	186.8 _{abc}	16.7	685.0 _a	36.1	611.4 _a	18.5	319.8 _a	26.4	45.4 _a	6.3	158.6 _{abc}	20.8
174	147.9 _c	34.3	637.7 _{abc}	16.8	630.0 _a	11.1	337.8 _a	3.3	46.4 _a	1.4	151.4 _{abcd}	5.8
411	159.8 _{abc}	12.6	655.7 _{abc}	34.4	639.5 _a	27.9	339.3 _a	25.0	40.6 _a	2.3	136.8 _{bcd}	21.2
590	159.0 _{abc}	21.1	629.6 _{abc}	27.0	643.1 _a	30.9	358.9 _a	29.4	44.2 _a	1.1	130.4 _d	11.8
651	167.1 _{abc}	7.2	652.8 _{abc}	14.7	617.1 _a	28.8	332.2 _a	38.7	46.7 _a	2.1	140.8 _{abcd}	18.5
670	165.6 _{abc}	11.6	626.0 _{abc}	31.0	609.0 _a	33.3	342.4 _a	23.7	53.5 _a	7.0	156.6 _{abcd}	23.8
768	151.7 _{abc}	7.5	690.2 _a	40.2	605.3 _a	14.2	311.7 _a	12.5	42.9 _a	1.9	148.7 _{abcd}	5.9
776	151.4 _{abc}	3.3	647.9 _{abc}	28.8	617.9 _a	13.6	346.3 _a	16.8	45.2 _a	3.1	144.0 _{abcd}	26.1
844	173.5 _{abc}	13.0	593.3 _{bc}	17.7	616.8 _a	15.9	324.7 _a	10.9	47.6 _a	5.0	149.4 _{abcd}	17.1
859	202.6 _a	31.4	577.5 _c	15.8	608.2 _a	46.9	329.8 _a	19.8	47.3 _a	5.0	153.7 _{abcd}	22.7
950	206.3 _{ab}	8.9	638.7 _{abc}	7.0	590.2 _a	12.6	300.0 _a	1.8	49.0 _a	13.8	168.3 _{abcd}	19.3
965	172.7 _{abc}	3.7	667.1 _{ab}	15.3	616.0 _a	8.0	319.3 _a	14.3	39.0 _a	2.5	148.1 _{abcd}	14.6
970	180.7 _{abc}	1.9	679.7 _a	31.9	602.1 _a	18.7	313.4 _a	13.7	46.1 _a	0.5	155.4 _{abcd}	12.4
975	148.2 _{bc}	7.7	640.2 _{abc}	18.5	635.3 _a	28.6	336.7 _a	28.3	42.1 _a	7.8	138.8 _{abcd}	7.8
1067	184.1 _{abc}	27.3	683.7 _a	68.3	623.7 _a	51.1	331.2 _a	58.8	43.6 _a	5.1	145.8 _{abcd}	29.7
1093	166.2 _{abc}	8.8	638.2 _{abc}	15.9	630.9 _a	27.6	340.9 _a	24.4	52.0 _a	3.3	160.4 _{ab}	23.4
1296	148.4 _{bc}	21.9	640.8 _{abc}	29.0	637.1 _a	33.2	354.6 _a	27.9	45.2 _a	3.5	143.3 _{abcd}	27.4
1765	163.3 _{abc}	6.3	647.5 _{abc}	43.7	631.9 _a	39.0	334.1 _a	31.4	45.1 _a	1.8	149.5 _{abcd}	22.5
1806	147.0 _c	10.5	637.8 _{abc}	16.4	618.5 _a	24.0	320.4 _a	24.5	47.7 _a	4.6	154.5 _{abcd}	14.3
1894	172.2 _{abc}	24.7	651.6 _{abc}	36.6	624.7 _a	21.5	331.5 _a	28.1	42.2 _a	7.6	141.4 _{abcd}	25.3
1972	160.2 _{abc}	21.7	644.9 _{abc}	12.1	628.9 _a	23.5	347.8 _a	25.1	48.9 _a	0.1	132.4 _{cd}	17.4
<i>Ruziziensis</i>	151.1 _{abc}	6.9	633.9 _{abc}	24.8	618.9 _a	20.4	343.6 _a	23.7	44.1 _a	5.7	141.8 _{abcd}	26.0
<i>Brizantha</i>	173.7 _{abc}	14.5	629.2 _{abc}	19.0	631.6 _a	13.1	342.8 _a	21.1	48.2 _a	2.9	150.4 _{abcd}	12.8
<i>Decumbens</i>	187.9 _{abc}	17.5	649.8 _{abc}	14.0	628.7 _a	28.9	334.8 _a	21.4	42.7 _a	3.7	137.7 _{bcd}	10.1
Average	166.9		646.9		620.1		331.3		45.5		148.5	
SD	20.7		35.1		25.8		25.8		5.0		18.8	
<i>P-value</i>	0.001		0.000		0.110		0.016		0.123		0.000	

SD = Standard deviation of the mean; Means followed by the same letter within Column, do not differ by Tukey test ($P < 0.05$).

No difference ($P>0.05$) was observed in neutral detergent fiber (NDF), acid detergent fiber (ADF) and lignin (Lig). The mean value of Lig was 45.5g/kg DM. For the NDF and ADF, the variations were 643.1 to 580.4 and 358.9 to 288.5g/kg of DM, respectively. According to VAN SOEST (1994), the NDF content influences the consumption of bulky foods and all clones presented values higher than 550g/kg, considered by this author as a limit to influence the consumption of forage.

LOPES et al. (2010) evaluated the nutritional quality of four *Brachiaria* species (*B. brizantha*, *B. humidicula*, *B. decumbens* and *B. ruziziensis*) at 56 days of growth and reported mean values of DM, NDF and ADF higher than those observed in the present

study, 20.55, 68.44 and 36.53%, respectively, while mean values of CP, IVDMD and Lig were lower (6.9, 61.6 and 3.3%, respectively). Higher values of NDF and ADF and lower values of IVDMD and CP were also observed by SOUZA SOBRINHO et al. (2011) (NDF, ADF, IVDMD and CP of 78.0, 43.1, 55.2 and 6.3%, respectively) were evaluated for the forage quality of different species of *Brachiaria*, cut at 57 days of regrowth. Difference observed between the studies can be attributed to the stage of forage maturity, since digestibility and CP tend to decrease with the advancement of plant maturity and increase of the NDF and ADF fraction.

Table 2 shows the parameters of ruminal fermentation kinetics of *B. ruziziensis* clones and controls.

Table 2 - Average of the adjusted parameters in relation to the *in vitro* fermentation kinetics of the fibrous carbohydrates (FC) and non-fibrous carbohydrates (NFC) in relation to the *Brachiaria ruziziensis* progenies and the *Brachiaria ruziziensis* cv. 'Basilisk'.

Clones	Variables									
	VFNFC (ml/g ⁻¹)	SE	KdNFC (h ⁻¹)	SE	L (h:min)	SE	VFFC (ml/g ⁻¹)	SE	KdFC (h ⁻¹)	SE
15	57.8	18.6	0.068	0.02	5:24	0.6	134.8	17.8	0.025	0.00
16	62.3	23.2	0.059	0.01	5:30	0.6	133.0	22.2	0.023	0.00
46	92.8	28.3	0.051	0.01	4:47	0.7	123.7	26.9	0.020	0.00
174	118.7	19.9	0.042	0.00	8:12	0.5	79.3	17.6	0.015	0.00
411	85.2	22.8	0.053	0.01	6:07	0.1	126.8	21.6	0.020	0.00
590	92.7	24.4	0.048	0.01	6:28	0.5	115.4	23.2	0.020	0.00
651	49.7	18.8	0.063	0.02	5:33	0.7	142.0	17.8	0.022	0.00
670	73.6	22.5	0.054	0.01	5:15	0.7	114.9	21.4	0.021	0.00
768	86.9	20.6	0.053	0.01	6:12	0.5	98.5	19.7	0.021	0.00
776	11.9	19.4	0.040	0.00	9:04	0.4	66.1	17.1	0.015	0.00
844	85.6	26.9	0.037	0.01	8:33	0.6	69.3	24.3	0.015	0.00
859	93.7	17.3	0.049	0.01	6:10	0.4	80.3	16.3	0.019	0.00
950	61.7	15.7	0.064	0.01	3:55	0.6	125.9	15.0	0.023	0.00
965	49.4	13.1	0.088	0.02	4:58	0.7	170.2	12.6	0.025	0.00
970	42.8	11.5	0.091	0.02	5:35	0.7	165.9	11.1	0.025	0.00
975	101.5	29.4	0.041	0.01	6:06	0.5	87.8	27.7	0.018	0.00
1067	88.4	36.9	0.043	0.01	6:44	0.8	75.5	34.9	0.018	0.01
1093	79.0	21.4	0.042	0.01	6:18	0.6	74.7	19.7	0.017	0.00
1296	122.9	28.9	0.036	0.00	6:27	0.6	60.9	25.4	0.014	0.01
1765	111.2	37.8	0.036	0.01	6:15	0.7	75.7	34.6	0.016	0.01
1806	60.1	16.4	0.074	0.02	5:08	0.6	135.2	15.7	0.025	0.00
1894	83.5	21.0	0.050	0.01	4:25	0.6	119.4	19.8	0.019	0.00
1972	125.4	21.5	0.038	0.00	5:47	0.5	60.7	19.1	0.015	0.00
<i>B. ruziziensis</i>	91.3	32.7	0.044	0.01	5:56	0.7	108.2	31.0	0.019	0.00
<i>B. brizantha</i>	91.9	22.2	0.046	0.01	5:39	0.6	95.4	20.6	0.018	0.00
<i>B. decumbens</i>	87.4	21.8	0.048	0.01	6:55	0.6	105.4	20.3	0.018	0.00
Average	85.0		0.052		6:03		105.6		0.019	

VFNFC = equivalent to the maximum volume of gases from the NFC fraction (ml/g⁻¹); kdNFC = the degradation rate (h⁻¹) of NFC; VFFC = maximum volume of the gases from the FC fraction (ml/g⁻¹); kdFC = degradation rate (h⁻¹) of FC; L = lag time (hours:minutes); V_t = total gas accumulated at time t and SE = Standard error.

Cumulative gas production rate for the fermentation of fibrous and non-fibrous carbohydrates (VFFC and VFNFC) ranged from 60.9 to 170.2mL/g and from 42.8 to 125.4mL/g, respectively. Volume of gas produced depends on the composition of the food and, the larger the amount of fiber, the greater the gas production (NOGUEIRA et al., 2006). This allowed that genotypes that presented superior VFFC are more digestible compared to the fibrous fraction. Structural carbohydrates have slower degradability and for this reason, the fibrous carbohydrate degradation rate (KdFC) is lower than the non-fibrous carbohydrate degradation rate (KdNFC), 0.019 and 0.052DM/h, respectively.

The estimated colonization time (L) presented an average value of 6h. This parameter indicated the time involved between the beginning of the incubation and the start of microbial action on the sample. Thus, the greater amount of readily fermentable substances and the physical and chemical characteristics of the sample's cell wall, which facilitated microbial colonization, represented lower time of colonization (MAGALHÃES et al., 2006).

Assessing the kinetic parameters of ruminal degradation of the fibrous and non-fibrous carbohydrate fractions of *Brachiaria brizantha* cv. 'Marandu' at three ages of cuts (28, 35 and 54 days) by the *in vitro* technique of gas production, SÁ et al. (2011), reported mean values for VFNFC and KdNFC at 28 days of age of 93.51mL/g of DM and 0.05h⁻¹, respectively, values similar to those reported in the present study. However, the mean values

observed for VFFC and KdFC (83.25mL/g DM and 0.01DM/h⁻¹) were lower than those observed in the present study.

The evaluation of the nutritional divergence of *Brachiaria* genotypes (Table 3), was based on a principal component analysis (PC) where the cumulative variance of the first two principal components (PC1: 70.16% and PC2: 26.08%) explained 96.24% of variance between genotypes. Initially, we used all the variables of chemical and kinetic composition of fermentation, taken from minor models for discrimination of genotypes.

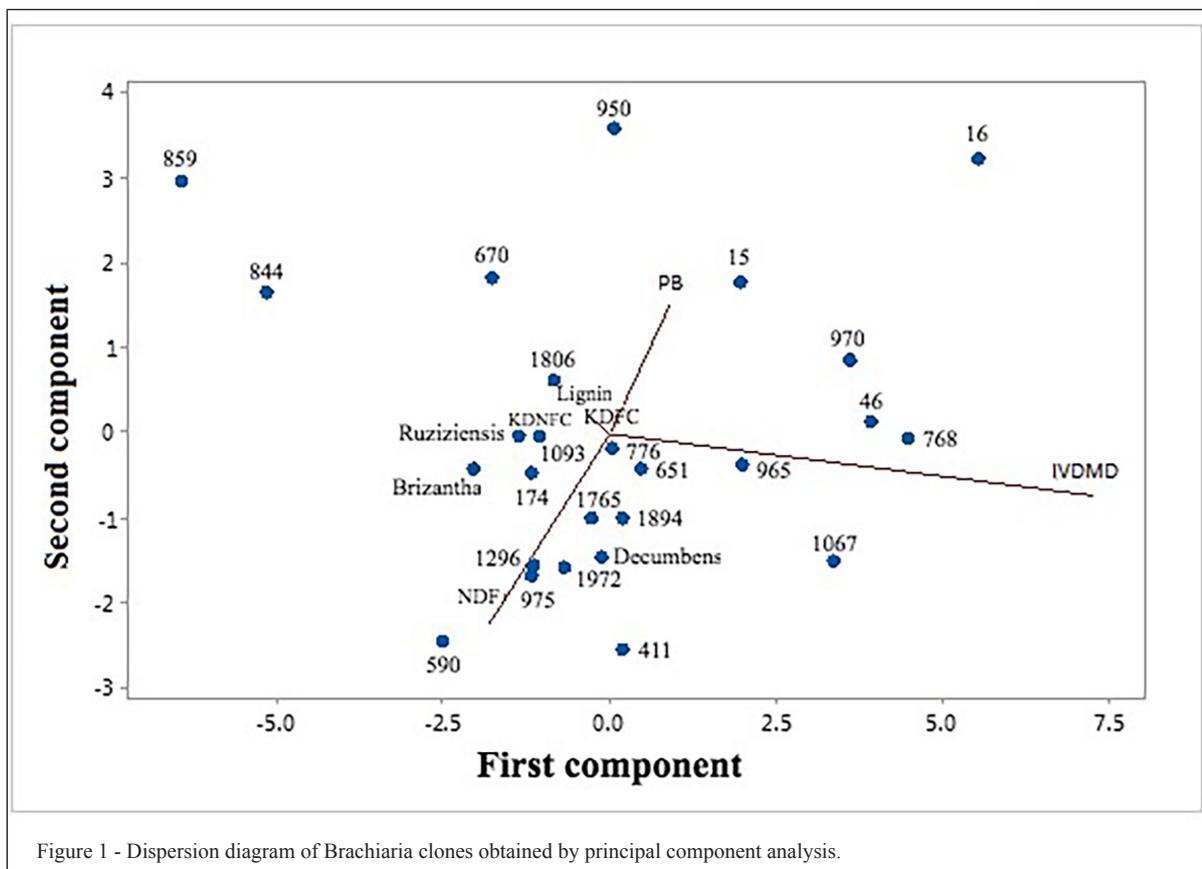
According to CRUZ et al. (2012), the relative importance of the main components decreases from the first to the last; the last component is responsible for explaining a tiny fraction of the total variance available. Thus, it was reported that the variables of lesser interest were KdNFC and KdFC once had a higher weighting in the smallest eigenvalue component (Table 3).

From the eigenvectors associated with the main components, we obtained the scores of the 26 *Brachiaria* genotypes. Graphical dispersion of the scores of the two main components can be reported in figure 1, where the distance of these points is proportional to the degree of dissimilarity between populations. Genotype grouping was observed in four sets. The first with clones 46, 768, 965, 970 and 106, the second one with clones 15, 16 and 950, the third one with clones 670, 844 and 859 and finally the fourth set with clones 174, 411, 590, 651, 776, 975, 1093, 1296, 1765, 1806, 1894, 1972 and the *Brizantha*, *Decumbens* and

Table 3 - Estimates of the eigenvalues of the cumulative variance and weighting of variables of the characters in the principal components obtained based on six variables in 26 genotypes of *Brachiaria*.

Principal Component	Variance (eigen value)	Percentage of Total Variance (%)	Variance Cumulative (%)	-----Weighting of variable-----					
				DIVDM	NDF	Lignin	CP	KdNFC	KdFC
1	7.5142	70.16	70.16	0.9620	-0.2400	-0.0476	0.1213	0.0023	0.0006
2	2.7933	26.08	96.24	0.2627	0.7973	-0.0802	-0.5375	-0.0031	-0.0009
3	0.3389	3.16	99.40	0.0446	0.5514	0.1883	0.8115	-0.0037	-0.0008
4	0.0641	0.60	100.00	0.0598	-0.0525	0.9776	-0.1945	-0.0068	-0.0019
5	0.0002	0.00	100.00	-0.0009	0.0048	0.0074	-0.0003	0.9822	0.1874
6	0.0000	0.00	100.00	0.0000	0.0003	0.0006	-0.0002	-0.1874	0.9823

IVDMD = *in vitro* dry matter digestibility; NDF = neutral detergent fiber; CP = crude protein; kdNFC = degradation rate (h⁻¹) of NFC and kdFC = degradation rate (h⁻¹) of FC.



Ruziziensis controls. Clones 15, 16, 670, 844, 859 and 950 showed the highest dispersion of scores in the first two main components and were considered the most dissimilar.

For the hierarchical clustering analysis by full connection method, based on Euclidean distance average, we used six variables selected from the PC analysis (IVDMD, NDF, lignin, CP, KdNFC and KdFC) and obtained five distinct groups (Table 4). Group V, formed by clones 46, 768 and 1067, showed better results in relation to the others, with a higher average of IVDMD (686.3g/kg), which was the main discriminating factor. Group IV constituted by clones 15, 16 and 950 is distinguished by the low NDF, high CP contents and high degradation rate of the fibrous fraction.

Group V clones have IVDMD values higher than Group I and II that include traditional cultivars already consolidated in the domestic market, indicating the potential of the nutritional

value of the members of this group for the breeding program. Group I introduced many clones and also included *B. decumbens* and *B. ruziziensis* (Kennedy). Group II, consisting of clones 670, 844, 859, 1093 and *B. brizantha*, showed the lowest IVDMD (612.8g/kg) and higher lignin content (49.7g/kg). Clones of this group have similar features to *B. brizantha*, a species of great importance in Brazilian livestock rearing.

CONCLUSION

B. ruziziensis clones showed nutritional divergence and clones 46, 768 and 1067 were distinguished clones of high nutritional value. Clones 15, 16 and 950 are distinguished by the lower values of NDF and high protein levels. The divergent nutritional characteristics can guide new crosses in the breeding program complementing the agronomic parameters for the generation of superior genotypes.

Table 4 - Groups of genotypes of *Brachiaria* and average variables in each group formed by the hierarchical agglomerative clustering method of Complete Linkage, based on standardized average Euclidean distance.

Items	Group				
	I	II	III	IV	V
Clones					
	174	670	651	15	46
	411	844	965	16	768
	590	859	970	950	1067
	776	1093	1806		
	975	<i>B. brizantha</i>			
	<i>B. ruziziensis</i>				
	1296				
	1765				
	1894				
	1972				
	<i>B. decumbens</i>				
DIVDM (%DM)	643.6	612.8	659.4	664.1	686.3
NDF (%DM)	630.5	619.3	613.4	591.1	613.5
Lignin (% DM)	44.2	49.7	44.9	45.5	44.0
CP (%DM)	140.7	154.1	149.7	168.9	151.0
KdNFC (h ⁻¹)	0.043	0.045	0.079	0.063	0.049
KdFC (h ⁻¹)	0.017	0.018	0.024	0.024	0.020

IVDMD = *in vitro* dry matter digestibility; NDF = neutral detergent fiber; CP = crude protein; KdNFC = degradation rate (h⁻¹) of NFC and KdFC = degradation rate (h⁻¹) of FC.

ACKNOWLEDGEMENTS

The authors acknowledge the Embrapa Gado de Leite, Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG), Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq – Edital Repensa - Projeto PECUS-RumenGases) for the financial support and to Fundação de Amparo à Pesquisa do Estado da Bahia (FAPESB) for the granting scholarship.

BIOETHICS AND BIOSSECURITY COMMITTEE APPROVAL

All the management procedures of the animals were conducted according to ethical principles of animal experimentation, established by the Brazilian College of Animal Experimentation and the current legislation was approved by the Ethics Committee for Animal Use of EMBRAPA Dairy Cattle (No. 03/2014).

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