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Research article

Evaluation of sugarcane rind on the nutritional value of ruminant feeding

Raiany Resende Moura[1], Michele Gabriel Camilo[1], Elizabeth Fônseca Processi[2], Alberto Magno Fernandes[1], Ismael Nacarati da Silva[1], Elon Souza Aniceto[1], Tadeu Silva de Oliveira[1]*

[1]Universidade Estadual do Norte Fluminense Darcy Ribeiro – Lab. de Zootecnia, Av. Alberto Lamego, 2000 – 28013-602 – Campos dos Goytacazes, RJ – Brasil.
[2]Universidade Federal Rural do Rio de Janeiro – Campus Campos dos Goytacazes, Av. Lourival Martins Beda, s/n – 28022-560 – Campos dos Goytacazes, RJ – Brasil.
*Corresponding author <tsoliveira@uenf.br>

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ABSTRACT: Several studies on the kinetics of sugarcane's fiber digestion have been published, but, to date, no study has evaluated the influence of sugarcane rind on the digestion of fresh sugarcane by ruminants. This study aimed to evaluate the effects of sugarcane components (rind and pith) on chemical composition, *in vitro* digestibility, metabolizable energy, and sugarcane quality. A randomized block design was used in a split-plot scheme with five sugarcane genotypes [plot] (RB068027, RB058046, RB987917, RB867515, and RB855536) and three sugarcane components [sub-plot] (rind, pith, and whole cane), Each treatment consisted of four replicates. The chemical composition, *in vitro* gas production, *in vitro* digestibility, metabolizable energy, and sugarcane quality were evaluated. No interaction between components and genotypes was observed for the variables analyzed herein. Although the rind had a higher crude protein content, it showed a large amount of insoluble crude protein. The rind had higher fibrous fractions, comprising 87.33 % of the indigestible fraction of the neutral detergent fiber (NDF). The sugarcane rind showed ~ 71.20 % more lignin than the pith tissue. Further, the rind decreased by 6.5 % *in vitro* dry matter digestibility compared to the whole sugarcane. The *in vitro* NDF digestibility of the rind was 18.38 % lower than the whole sugarcane. The RB068027 genotype showed the lowest sugarcane quality. Despite the higher content of potentially digestible neutral detergent fiber (pdNDF) in the rind, its high lignin content influences the quality of the final fibrous fractions of sugarcane and negatively impacts the nutritional value. The genotypes do not differ nutritionally, but RB855536 presented higher biomass and energy yields.
Keywords: *Saccharum officinarum* L., digestion, fibrous fractions, forage

Introduction

Sugarcane is grown extensively in America, Africa, Asia, and Oceania on account of its ease of cultivation and outstanding production of green mass, which facilitate its use in ruminant feeding during the dry season (characterized by low rainfall and high temperatures), which results in a shortage of forage (Bento et al., 2018). Thus, sugarcane is an excellent option for farmers as it has advantages such as great nutritional value and forage production per area, concurring with the period of forage shortage compared to tropical forages (Gomes et al., 2016). However, sugarcane production in Brazil focuses on the sugar-energy industry rather than on animal nutrition, making selecting varieties with better nutritional value for animal feeding necessary (Carvalho et al., 2022).

Sugarcane deserves some attention due to its nutritional limitations, such as low protein and mineral levels and low-quality fibrous fractions. Among the nutritional limitations, the fibrous fractions significantly impact feed digestion, and protein and minerals can be corrected by supplements (Gomes et al., 2016). Sugarcane constituents with high lignification are in the strongly recalcitrant rind (Maziero et al., 2013). The tissue architecture of lignin and the suberin lamellae's aromatic fraction may be a significant physicochemical factor limiting rumen microorganisms' degradation of sugarcane (Maziero et al., 2013).

Moreover, the existence of a genetic variability effect between sugarcane genotypes on fibrous fraction is a possibility. Fiber-related traits were neglected by selection, resulting in cultivars having more genetic variability available for fiber-related than for sugar-related traits (Cursi et al., 2021). There are many studies on sugarcane's chemical composition and fiber digestion kinetics, but to date, no study has evaluated the influence of the rind on the digestion of fresh sugarcane. In addition to mechanical protection and water retention, the rind can drain the stored carbon into the stalk, which accumulates sucrose (Wang et al., 2013). Based on the above, it was hypothesized that: 1) the fibrous fractions of the sugarcane pith would have as much impact on the nutritional value as the fibrous fractions of the rind; and 2) there would be a nutritional difference between genotypes. Thus, the present study aimed to evaluate the effect of components (rind and pith) of five sugarcane genotypes on chemical composition, *in vitro* digestibility, metabolizable energy, and sugarcane quality.

Materials and Methods

Location

The experiment was conducted in Bom Jesus do Itabapoana, Rio de Janeiro, Brazil (21°08′13″ S, 41°39′30″ W, 85 m altitude). The climate of the northern

state of Rio de Janeiro is partial Aw, i.e., humid tropical with rainy summers and dry winters, with an average annual temperature of 23 °C and rainfall of 1,200 mm according to the Köppen-Geiger classification (Alvares et al., 2013).

Area and experimental design

The soil was prepared with plowing, harrowing, and furrowing, according to Portz et al. (2013). Before planting, some soil samples were sent for analysis at the Analysis Center of the Universidade Federal Rural do Rio de Janeiro, Campos dos Goytacazes, Rio de Janeiro. The soil presented the following chemical composition: pH in H_2O = 5.8 P (mehlich) = 8 mg dm^{-3}; K = 21.5 mg dm^{-3}; Na = 0.0 mg dm^{-3}; Ca = 1.5 cmol dm^{-3} ; Mg = 0.7 cmol dm^{-3}; Al = 0.0 cmol dm^{-3}; H + Al = 2.7 cmol dm^{-3}; CEC (t) = 1.1 cmol dm^{-3}; CEC (T) = 2.2 cmol dm^{-3}; SB = 2.2 cmol dm^{-3}; BS = 45.6 %; OM = 1.7 %; Fe = 59.5 mg dm^{-3}; Cu = 0.3 mg dm^{-3}; Zn = 300.6 mg dm^{-3}; and Mn = 18.6 mg dm^{-3}. The area was fertilized following the recommendations of the Liming and Fertilization Manual of the State of Rio de Janeiro for sugarcane crops (Portz et al., 2013). Four hundred kg ha^{-1} of formulated NPK 08-28-16 was applied. In order to reduce area heterogeneity [soil fertility]) the randomized block used a split-plot design. The factors were arranged per the experimental design as main factor plot genotypes and sub-plot factor components of sugarcane. There were five sugarcane genotypes from the Interuniversity Network for Development of the Sugar-Energy Sector (INDSES), with four replicates for each genotype. The genotypes were RB867515 [G1], RB855536 [G2], RB068027 [G3], RB058046 [G4], and RB987917 [G5]. Three components of sugarcane (rind, pith, and whole cane) were evaluated. Each replicate had four lines, 4 m long, and a spacing of 1.20 m, totaling 19.2 m^2 of useful area per replicate.

The harvest was carried out in Aug 2020. Ten whole sugarcanes were harvested from the third row of each plot, and their weight recorded. Next, five stalks were taken for sugarcane quality testing. Five canes were rinded with a spoon so that all the pith in the rind was removed, and the other five canes had the aerial part (whole cane) stripped off.

Chemical composition

The sugarcane samples were taken to the Animal Nutrition Laboratory of the Universidade Estadual do Norte Fluminense (UENF) and separated into rind, pith, and whole sugarcane. They were dried at 55 °C for 72 h, ground in a Wiley mill (R-TE-648, Tecnal) with a 1-mm-sieve, and stored in airtight plastic containers. All samples were analyzed for total dry matter (DM, method 967.03; AOAC, 2019), crude fat (CF, AOAC Method 2003.06; Thiex et al., 2003), ash (method 942.05; AOAC, 2019), and crude protein ([N × 6.25] CP, AOAC Method 984.13 and AOAC Method 2001.11; AOAC, 2019; Thiex et al., 2002). Neutral detergent insoluble fiber (aNDF) was determined using the fiber analyzer (Tecnal TE-149). Sodium sulfite and two standardized heat-stable α-Amylase solution additions were used according to the INCT-CA method F-001/1, as described by Detmann et al. (2012). The acid detergent fiber (ADF) was analyzed also according to the INCT-CA-F-003/1 method described by Detmann et al. (2012) and the lignin (sa) content by Möller (2009). Non-fibrous carbohydrate (NFC) was estimated as NFC (g kg^{-1}) = 1000 – CP – CF – Ash – aNDF. The content of neutral detergent soluble (NDS) was obtained by subtracting NDS = 1000 – aNDF. Neutral detergent insoluble crude protein (NDICP) was determined by analyzing the aNDF residues for Kjeldahl nitrogen (Licitra et al., 1996).

For the analysis of indigestible neutral detergent fiber (iNDF), the rind, pith, and whole sugarcane were processed in a Wiley mill with a 4-mm-sieve and stored in 13 × 7 cm nylon bags, 50 µm of pore diameter, a ratio of 25 mg of DM cm^{-2} of the bags' surface. The bags with samples were tied on a steel chain with a 250 g anchor and introduced into the rumen of four cannulated sheep for 240 h. Next, the material was taken from the rumen and washed under running water until there were no traces of ruminal residue. Subsequently, the samples were dried in a forced-air oven at ± 55 °C for 72 h, and the weight was determined on an analytical scale for further aNDF analysis according to the INCT-CA F-001 method /1, as described by Detmann et al. (2012).

Gas production kinetics, *in vitro* digestibility, and metabolizable and net energy

Four cannulated sheep were used in this study. The Ethics Committee approved all experimental procedures on the Use of Experimental Animals, protocol 419/2017. The animals weighed 50 kg (standard deviation = 4.1 kg) and were used as donors of ruminal fluid. They were kept in collective stalls with troughs and drinkers. Before ruminal fluid collections, the sheep were adapted to a diet of Tifton 85 hay and concentrate feed (roughage:concentrate ratio [80:20]) with 100 g d^{-1} of sugar for 14 days. After this period, the ruminal fluid collections were initiated moments before daytime feeding, as Yáñez-Ruiz et al. (2016) recommended.

The ruminal fluid (liquid and solid) was collected at several points on the liquid-solid interface of the ruminal environment via cannula using a collecting cup. A buffer solution described by McDougall (1948) was added. Two hundred mg (standard deviation = 10 mg) of rind, pith, and whole sugarcane samples from the five sugarcane genotypes were added in amber penicillin flasks (100 mL) with 20 mL of the previously prepared inoculum [ratio 1:4; ruminal fluid and buffer solution, respectively, according to Goering and Van Soest (1970)]. The free space in the

flasks was immediately saturated with CO_2. Next, the flasks were sealed and taken to a water bath at 39 °C, where they were shaken during incubation to homogenize the inner content. *In vitro* incubations were conducted in two consecutive runs, each with the sample in triplicates.

Time profiles of accumulated gas production were obtained using a non-automated device. A 0 to 8 psi manometer (0.05 increments) was attached to a three-way plastic valve. One of the ways was connected to a silicone tube (i.d. 5 mm; 1.5 m in length) with a 20 gauge needle attached to the loose extremity of the tube. The second way was attached to the manometer by a small piece of the silicone tube (i.d. 5 mm; 0.3 m in length) and plastic clamps. The third way was connected by another silicone tube (i.d. 5 mm; 1.3 m in length) to the top of a graduated 25 mL pipette (0.1 mL increments), which had its conical end connected to the stem of a separating funnel (1,000 mL) by the same type of silicone tube (i.d. 5 mm; 0.4 m in length). The funnel and pipette were attached to a metal support stand in a vertical and static position. The connecting system was filled with resazurin solution (0.1 g L^{-1}) to the zero mark of the pipette, i.e., it allowed for atmospheric pressure equilibration. The system was cautiously filled to avoid the formation of air bubbles. Pressure and volume were taken at: 0, 1, 2, 3, 4, 6, 8, 10, 12, 16, 20, 24, 30, 36, and 48 h after the ruminal inoculum was added. The pressure and cumulative volume of the fermentation gases were obtained by summing the readings corrected to the mark after zero.

The model used to estimate the cumulative gas production was proposed by Groot et al. (1996):

$$G = A/(1 + (B^c/t^c)) \quad (1)$$

$$R_M\,(\text{mL h}^{-1}) = ((C \times t^{(C-1)}) / (B^C + t^C)) \quad (2)$$

where: G is the amount of gas produced per unit of dry matter (DM) at time t after the incubation started, A, the asymptotic gas production (mg g^{-1} DM), B, the time (h) after incubation in which half of the asymptotic gas was formed representing the speed of gas production, C, a constant that determines the sharpness of the curve change; and R_M the maximum gas production rate when the microbial population does not limit the fermentation and digestion is not reduced by chemical or structural barriers of the potentially digestible material.

The determination of *in vitro* digestibility was focused on a single digestion step of the ruminal fluid, omitting the step with pepsin recommended by Tilley and Terry (1963). The buffer solution was the same as mentioned above. For each sample (rind, pith, and whole cane), triplicates of approximately 200 mg of air-dried samples were weighed and placed in 100 mL amber penicillin flasks with 20 mL of buffer solution and inoculum. The free space in the flasks was immediately saturated with CO_2, sealed, and taken to a water bath at 39 °C.

After 48 h of incubation, the flasks were withdrawn from the water bath, washed with hot distilled water (above 90 °C), and the incubated material filtered through quantitative filter paper (55 L s^{-1} m^2 air permeability). The resulting material was dried (55 °C 24 h^{-1} followed by 105 °C 16 h^{-1}) and weighed to obtain the apparently undigested residue of dry matter (DM). Next, that material was analyzed for *in vitro* digestibility of NDF implementing the methodology described by Detmann et al. (2012). The potentially digestible fraction was determined by subtracting NDF from iNDF.

The digestibility (D) of DM and NDF was calculated according to the Eq. (3):

$$D = (M - [R - B]/M) \times 1000 \quad (3)$$

where: M = mass of DM or NDF incubated (g); R = DM or NDF residue from incubation (g); B = DM or NDF residue of the blanks (g).

Metabolizable energy (ME) and net energy (NE) of the rind, pith, and whole sugarcane of the five genotypes were estimated using the equations by Menke and Steingass (1988):

$$\text{ME, MJ kg}^{-1}\,\text{DM} = 0.157 \times \text{GP} + 0.0084 \times \text{CP} + 0.022 \times \text{CF} - 0.0081 \times \text{Ash} + 1.06 \quad (4)$$

$$\text{NE, MJ kg}^{-1}\,\text{DM} = 0.115 \times \text{GP} + 0.0054 \times \text{CP} + 0.014 \times \text{CF} - 0.0054 \times \text{Ash} + 0.36 \quad (5)$$

where: GP is the net gas production over 24 h (mL mg^{-1} DM).

Sugarcane quality

Five culms from each experimental plot were taken to the Coagro (Cooperativa Agroindustrial do Estado do Rio de Janeiro Ltda.) to conduct the technological analyses according to the methodologies suggested by CONSECANA (2006). Technological analyses were performed only for the whole sugarcane.

The automatic hydraulic press method performed the brix and polarization of sugarcane (POL) analyses from the juice (Codistil). Brix (%) was analyzed using a digital refractometer (Acatec RDA8600) with automatic reading and corrected temperature. An automatic saccharimeter was used to determine the POL (Acatec DAS2500). It was calibrated at 20 °C with a wavelength between 587 and 589.4 nm and fitted with a continuous flow polarimetric tube. The percentage of POL was obtained by the following Eq. (6):

$$POL, \% = (((1.0078 \times sacc.) + 0.0444) \times ((0.2607 - (0.009882 \times \% Brix)) \quad (6)$$

The apparent purity of the juice was obtained by the ratio between POL and Brix, according to Eq. (7):

$$Purity, \% = POL/Brix \times 100 \quad (7)$$

Total recoverable sugars (TRS) were determined based on Eq. (8):

$$TRS, \text{ t ha}^{-1} = ((10 \times S - 0.76 \times IF - 6.9) \times (5/3 - 200/3 \times P)) \quad (8)$$

where S is sucrose (%), and P represents the purity calculated by the ratio between % POL and % Brix.

The reducing sugars were calculated according to Eq. (9):

$$RS, \% = 3.641 - 0.0343 \times P \quad (9)$$

Equations (6), (7), (8), and (9) were proposed by CONSECANA (2006).

At the end of the cycle (360 days), the sugarcane was harvested and weighed by cutting two linear meters of the second line of each experimental replicate. Next, the weights of total biomass (stalks, leaves, and straw) were recorded and used to estimate the tons of stems per hectare (TSH) and corrected for DM content, expressed in t ha^{-1} of DM.

Statistical analysis

The chemical composition, cumulative gas production, metabolizable and net energy estimates, and *in vitro* digestibility were compared by Tukey test at 0.05 significance using the SAS MIXED package using REPEATED statement and option = SUBJECT = Block × genotypes for analyzing split-plot design (SAS OnDemand Academics, SAS Institute Inc.). No interaction was observed between genotypes and components in the analyzed variables. Thus, the genotype was tested with residue (a [numerator degree of freedom = 4 and denominator degree of freedom = 42]) and the components with residue (b [numerator degree of freedom = 2 and denominator degree of freedom = 42]).

The following statistical model was used:

$$Y_{ijk} = \mu + \alpha_i + b_k + \alpha b_{ik} + \beta_j + \alpha\beta_{ij} + e_{ijk}$$

where Y_{ijk} is the value observed for the variable under study referring to the *k-th* replicate of the *i-th* sugarcane genotype in the *j-th* component (whole sugarcane without aerial part; sugarcane without rind, and only the rind); μ, the mean of all experimental units for the variable under study; α_i the effect of the sugarcane genotype with i = 1,2,3,4,5; b_k the random effect of the *k-th* block on the observation, αb_{ik} the residue (a) associated with the plot; β_j the effect of the component with i = 1,2,3; $\alpha\beta_{ij,}$ the interaction between sugarcane genotypes and components, and $e_{ijk,}$ the residue (b) associated with the split-plots.

Sugarcane biomass and quality were compared by Tukey test at 0.05 significance using the SAS GLM package (SAS OnDemand Academics, SAS Institute Inc.). The following statistical model was used:

$$Y_{ij} = \mu + \alpha_i + b_j + e_{ij}$$

where Y_{ij} is the value observed for the variable under study referring to the *k-th* replicate in the *i-th* sugarcane genotype, μ, the mean of all experimental units for the variable under study, $\alpha_{i,}$ the effect of the sugarcane genotype with i = 1,2,3,4, $b_{j,}$ the random effect of the *j-th* block on the observation, and $e_{ij,}$ the error associated with observation Y_{ij}.

Results

Chemical composition

There was no interactive effect ($p \geq 0.05$) between components and genotypes on chemical composition (Tables 1 and 2). In the genotypes, G4 presented lower contents for CP ($p = 0.042$) and CF ($p = 0.035$) than G2, but it did not differ from the others (Tables 1 and 2). As for the components, although the rind had a higher CP content ($p < 0.001$), it had a large amount of NDICP ($p < 0.001$), approximately 34.55 % (10.19/15.57) more

Table 1 – *p*-values related to the measured variables analyzed for the effects of the genotypes, components, and genotypes by components interaction.

Variables	Genotypes	Components	Interaction
DM	0.143	< 0.0001	0.228
CP	0.042	< 0.0001	0.082
NDICP	0.714	< 0.0001	0.704
CF	0.035	< 0.0001	0.063
Ash	0.920	0.002	0.231
NFC	0.099	< 0.0001	0.417
NDF	0.147	< 0.0001	0.291
NDS	0.147	< 0.0001	0.291
ADF	0.317	< 0.0001	0.371
Lig	0.295	< 0.0001	0.489
iDM	0.184	< 0.0001	0.835
iNDF	0.188	< 0.0001	0.166
pdNDF	0.194	< 0.0001	0.714
Gas 24 h	0.228	0.001	0.591
ME	0.078	0.085	0.632
NE	0.093	0.091	0.625
IVDMD	0.610	< 0.0001	0.470
IVNDFD	0.231	< 0.0001	0.641

DM = Dry matter; CP = Crude protein; NDICP = Neutral detergent insoluble crude protein; CF = Crude fat; NFC = Non-fibrous carbohydrate; NDF = Neutral detergent fiber; NDS = Neutral detergent soluble; ADF = Acid detergent fiber; Lig = Lignin; iDM = Indigestible dry matter; iNDF = Indigestible neutral detergent fiber; pdNDF = potentially digestible neutral detergent fiber; Gas 24 h = Gas production in 24 h; ME = Metabolizable energy; NE = Net energy; IVDMD = *In vitro* dry matter digestibility; and IVNDFD = *In vitro* neutral detergent fiber digestibility.

than the whole sugarcane and 55.10 % more than the pith. The CF content was also higher for the rind than the whole sugarcane ($p < 0.001$), approximately 33.88 % more and 72.54 % more than the pith (Tables 1 and 2). However, the rind had lower DM ($p < 0.001$) and NFC ($p < 0.001$) content compared to the pith. Ash contents for the rind were higher ($p = 0.003$) than the pith, but they did not differ from the whole sugarcane (Tables 1 and 2).

Fibrous fractions

There was no interaction effect ($p \geq 0.05$) between components and genotypes on fibrous fractions (Tables 1 and 3). The genotypes did not affect ($p \geq 0.05$) the fibrous fractions of sugarcane (Tables 1 and 3). However, regarding the components, the rind impacted these fractions, presenting 34.52 % more NDF than whole sugarcane, of which 87.33 % corresponds to the indigestible fraction of NDF (Tables 1 and 3). Furthermore, the potentially digestible fraction of the rind was higher by 52.71 % than whole sugarcane and by 52.68 % more than pith. On the other hand, the NDS contents of the rind were higher ($p < 0.001$) by 42.21 % than the pith (Tables 1 and 3). Contents of ADF ($p < 0.001$) and indigestible dry matter (iDM) ($p < 0.001$) had similar behavior to NDF for the rind, pith, and whole sugarcane. As regards lignin contents, the rind was 71.20 % higher than the pith ($p = 0.002$) (Tables 1 and 3). The average values of the components for neutral detergent fiber content, indigestible neutral detergent fiber content, potentially digestible neutral detergent fiber content, and in vitro neutral detergent fiber digestibility are presented in Figures 1A-D.

Table 2 – Effects of components and genotypes on the chemical composition of sugarcane.

Variables	G1	G2	G3	G4	G5	SEM
	Genotypes					
DM	380.3	419.8	396.7	411.7	399.5	2.533
CP	17.4^{abc}	19.8^{a}	17.4^{abc}	14.0^{c}	15.7^{abc}	0.360
NDICP	11.1	11.9	11.2	9.2	11.3	0.154
CF	7.6^{abc}	8.8^{a}	6.8^{abc}	5.4^{c}	6.3^{abc}	0.221
Ashes	1.7	1.6	1.5	1.7	1.6	0.013
NFC	502.4	522.2	508.7	542.2	541.1	3.277
	Components					
	Rind		Pith		WC	
DM	483.38^{a}		326.95^{c}		394.45^{b}	12.193
CP	19.90^{a}		12.98^{c}		17.71^{b}	0.579
NDICP	15.56^{a}		6.99^{c}		10.18^{b}	0.694
CF	10.75^{a}		2.99^{c}		7.20^{b}	0.594
Ashes	1.85^{a}		1.12^{b}		1.84^{a}	0.064
NFC	373.09^{c}		682.79^{a}		514.05^{b}	23.774

DM = Dry matter as fed; CP = Crude protein; NDICP = Neutral detergent insoluble crude protein; CF = Crude fat; Ashes; and NFC = Non-fibrous carbohydrate. All expressed as g kg^{-1}. SEM = Standard error of the mean. WC = Whole cane. Genotypes: G1 = RB867515; G2 = RB855536; G3 = RB068027; G4 = RB058046; and G5 = RB987917. Means followed by the different letters differ significantly by the Tukey test ($p < 0.05$).

Gas production kinetics, *in vitro* digestibility, and metabolizable and net energy

There was no effect of interaction ($p \geq 0.05$) between components and genotypes on gas production, *in vitro* digestibility, and energy (Tables 1 and 4). The genotypes did not affect ($p \geq 0.05$) the gas production, *in vitro* dry matter digestibility (IVDMD), *in vitro* neutral detergent fiber digestibility (IVNDFD), nor

Table 3 – Effects of components and genotypes on the fibrous fractions of sugarcane.

Variables	G1	G2	G3	G4	G5	SEM
	Genotypes					
NDF	470.8	450.0	465.5	436.8	435.4	5.885
NDS	529.2	552.4	534.5	563.2	564.6	6.057
ADF	277.1	258.5	278.2	254.9	258.5	4.368
Lig	23.0	21.6	21.0	19.4	21.3	0.379
iDM	283.8	255.9	249.7	258.3	269.7	4.748
iNDF	421.1	404.7	409.9	383.0	392.8	5.152
pdNDF	49.7	42.9	55.6	53.8	53.8	1.739
	Components					
	Rind		Pith		WC	
NDF	595.47^{a}		300.0^{c}		459.6^{b}	58.385
NDS	404.54^{c}		700.0^{a}		541.8^{b}	58.204
ADF	380.59^{a}		155.84^{c}		259.9^{b}	44.320
Lig	34.73^{a}		10.0^{c}		19.0^{b}	5.191
iDM	391.13^{a}		152.49^{c}		246.9^{b}	49.122

NDF = Neutral detergent fiber; NDS = Neutral detergent soluble; ADF = Acid detergent fiber; Lig = Lignin; iDM = Indigestible dry matter; iNDF = Indigestible neutral detergent fiber; and pdNDF = potentially digestible neutral detergent fiber. All expressed as g kg^{-1}. SEM = Standard error of the mean. WC = Whole cane. Genotypes: G1 = RB867515; G2 = RB855536; G3 = RB068027; G4 = RB058046; and G5 = RB987917. Means followed by the different letters differ significantly by the Tukey test ($p < 0.05$).

Table 4 – Effects of components and genotypes on the gas production, energy, and *in vitro* digestibility of sugarcane.

Variables	G1	G2	G3	G4	G5	SEM
	Genotypes					
Gas 24 h	33.6	35.3	34.5	32.0	32.9	0.219
ME	7.1	7.5	7.2	6.6	6.8	0.060
NE	4.0	4.3	4.1	3.7	3.8	0.043
IVDMD	562.6	569.2	569.3	571.6	564.7	0.685
IVNDFD	358.4	365.1	380.8	389.4	361.7	2.509
	Components					
	Rind		Pith		WC	
Gas 24 h	31.40^{b}		37.19^{a}		32.4^{b}	0.527
ME	6.67		7.38		7.1	0.055
NE	3.69		4.22		4.0	0.040
IVDMD	542.81^{b}		579.04^{a}		580.5^{a}	3.672
IVNDFD	299.742^{c}		447.91^{a}		365.6^{b}	11.453

Gas 24 h = Gas production in 24 h (mg g^{-1} DM); ME = Metabolizable energy (MJ kg^{-1} DM); NE = Net energy (MJ kg^{-1} DM); IVDMD = In vitro dry matter digestibility (g kg^{-1}) and IVNDFD = *In vitro* neutral detergent fiber digestibility (g kg^{-1}). SEM = Standard error of the mean. WC = Whole cane. Genotypes: G1 = RB867515; G2 = RB855536; G3 = RB068027; G4 = RB058046; and G5 = RB987917. Means followed by the different letters differ significantly by the Tukey test ($p < 0.05$).

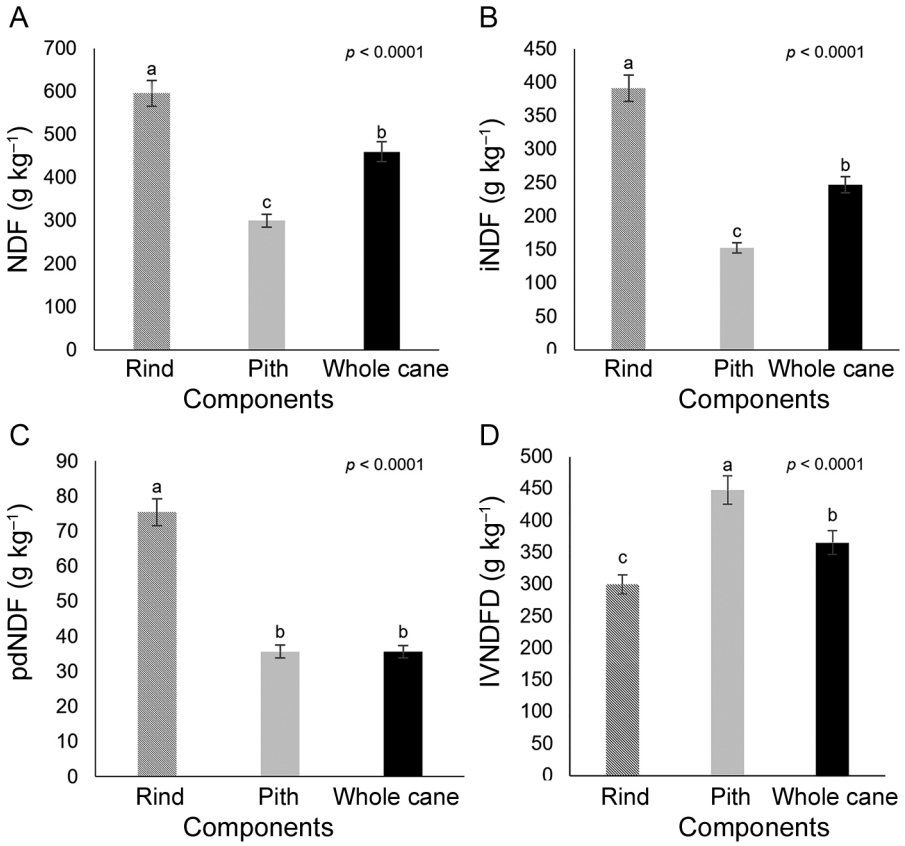

Figure 1 – Evaluation of the fibrous fractions of sugarcane components (rind, pith, and whole cane). A) NDF = Neutral detergent fiber; B) iNDF = Indigestible neutral detergent fiber; C) pdNDF = Potentially digestible neutral detergent fiber; and D) IVNDFD = *In vitro* neutral detergent fiber digestibility. All expressed in average values across genotypes.

the metabolizable and net energy of the sugarcane (Tables 1 and 4). When comparing the components, gas production for the rind was lower (p = 0.001) than that of the pith (Tables 1 and 4) within 24 h of *in vitro* incubation. However, there was no difference in components regarding sugarcane's metabolizable (p = 0.085) and net (p = 0.091) energy. Metabolizable and net energy for the rind were lower than the pith, approximately 9.61 and 12.38 % (Table 3), respectively. The rind presented IVDMD 6.5 % lower (p < 0.001) than the whole sugarcane. Additionally, IVNDFD was 18.38 % lower (365.59/447.91) (p < 0.001) than the whole sugarcane (Tables 1 and 4). There was no run effect (p = 0.526).

Sugarcane quality and biomass

Genotype did not affect on TSH (p = 0.173) or biomass (p = 0.771) (Table 5). However, G3 presented lower (p < 0.001) Brix than G1, G2, and G4. POL contents of the G3 genotype differed (p = 0.002) only in G1 and G5. Apparent purity was affected by genotypes (p < 0.001). G5 had 6.34 % more purity than G4. As regards TRS (p < 0.001) and RS (p < 0.001) sugars, only G2 did not differ from G3 (Table 5).

Table 5 – Effects of genotypes on the technological quality of sugarcane.

Variables	Genotypes					SEM	*p*-value
	G1	G2	G3	G4	G5		
Biomass	108.30	124.70	123.6	111.20	104.0	4.677	0.771
TSH	28.92	34.17	33.17	30.70	29.25	0.868	0.173
Brix, %	22.85^{a}	22.50^{a}	21.28^{b}	22.65^{a}	21.80^{ab}	0.113	< 0.001
POL	19.04^{a}	18.38^{ab}	17.38^{b}	18.33^{ab}	18.85^{a}	0.129	0.002
Purity, %	83.32^{a}	81.69^{ab}	81.67^{ab}	80.93^{b}	86.46^{a}	0.105	< 0.001
TRS	148.56^{a}	143.80^{ab}	137.50^{b}	149.20^{a}	149.85^{a}	0.778	< 0.001
RS	17.68^{a}	17.11^{ab}	16.36^{b}	17.75^{a}	17.83^{a}	0.093	< 0.001

Biomass (t ha^{-1}); TSH = Tons of stems per hectare (t ha^{-1} DM); POL = Polarization of sugarcane (%); TRS = Total recoverable sugars (kg t^{-1}); RS = Reducing sugars (%). SEM = Standard error of the mean. Genotypes: G1 = RB867515; G2 = RB855536; G3 = RB068027; G4 = RB058046; and G5 = RB987917. Means followed by the different letters differ significantly by the Tukey test (p < 0.05).

Discussion

Nutritional quality is essential in choosing a sugarcane variety for ruminant nutrition and productivity. However, one of the limitations of sugarcane in ruminant feeding is the low protein content and fiber digestibility. When

the industry selects varieties, little attention is paid to the variables of the plant that affect its nutritional value. This study observed a difference between genotypes for the CP content. G2 was higher (19.8 g kg^{-1}) than G4 (14.0 g kg^{-1}) (Tables 1 and 2). However, 67.82 % of the CP is in the rind, from which 81.34 % is in the form of NDICP (Tables 1 and 2). Neutral detergent insoluble crude protein represents the B3 fraction of protein fractioning, i.e., the fraction slowly degraded in the rumen because it adheres to the cell wall and is highly escapable from rumen degradation (Sniffen et al., 1992; Lanzas et al., 2008). The CF content was lower for G4 than for the other genotypes (Tables 1 and 2). However, the rind has higher CF content than the pith, and the sugarcane rind's wax, cutin, and suberin can explain this difference. They are polymerized fatty substances in the cell wall and reduce water loss from the plant (Nawrath, 2002). The wax of the rind has always been attractive for industrial applications, mainly in the cosmetic and pharmaceutical industries (Nuissier et al., 2002). The pith has lower DM than the rind (Tables 1 and 2). This fact is due to the thickness and impermeability of the fatty substances of the rind, thereby preventing water loss. Furthermore, the rind is structurally divided into the outer and inner rind. The outer rind comprises dead cells, providing structural support and protection against mechanical damage and pathogens. The inner rind comprises living tissue, including the phloem, responsible for storing and transporting water and solutes throughout the plant (Rosell et al., 2017). The higher NCF for the pith than the rind is due to the higher sucrose content in the pith. Ash had a higher content for the rind than the pith (Tables 1 and 2).

As regards the fibrous fractions, the rind showed higher levels of fiber than the pith. This result is due to the hemicellulose (NDF minus ADF), cellulose (ADF minus lignin), and lignin that grant greater rigidity, impermeability, and resistance to microbiological and mechanical attacks on plant tissues (Liu et al., 2018). It was observed that the indigestible fraction (iNDF) accounted for most NDF (Figures 1A and B). The indigestibility is probably related to the lignin of the rind and the compact organization of cellulose microfibrils in the hemicellulosic polysaccharide matrix covalently linked to a complex lignin structure (Vega-Sánchez and Ronald, 2010). Even so, lignin is the main component of the plant cell wall and is responsible for resistance to degradation (Bottcher et al., 2013). Thus, pdNDF presented low values, thereby reducing the fibrous fractions' availability to the ruminal microorganisms (Figure 1C). The iDM showed the same behavior as the iNDF (Tables 1 and 3). The lignin of the rind was 71.21 % higher than the pith. The low lignin content in the pith is due to the negative correlation between lignin and sucrose, which caused a dilution effect. Lignin drastically reduces the efficiency of saccharification in the pith since tissues rich in syringyl (S) are more susceptible to enzymatic hydrolysis than those rich in guaiacyl (G) (Bottcher et al., 2013). The most common monolignols for lignin polymer formation are *p*-hydroxyphenyl (H), guaiacyl (G), and syringyl (S) residues. They are secreted in the apoplast and deposited in the cell wall by extracellular peroxidases and laccases (Bottcher et al., 2013; Dixon and Barros, 2019; Llerena et al., 2019).

Cell wall digestibility is complex and can be influenced by several factors such as porosity, surface area, ratio of lignin monomers, cellulose crystallinity, degree of polymerization that limits the access of cellulolytic enzymes to cell wall polysaccharides, lignin, suberin, and cross-links with hemicellulose (Pu et al., 2013). In the present study, the rind presented lower IVNDFD content (Tables 1 and 4) than the pith, probably due to the higher contents of CF and lignin in the rind, thereby corroborating Pu et al. (2013). For Wilson and Mertens (1995), the rumen microorganisms cannot digest lignin and suberin (plant cell wall biopolymers). It prevents access to the polysaccharide matrix in the cell wall and affects digestibility. On the other hand, the pith showed higher IVNDFD content, which was caused by the higher saccharification, i.e., the number of sugar monomers released through enzymatic hydrolysis of cell wall polysaccharides (Ding et al., 2012). Unlike most grasses, the overall digestibility of sugarcane does not decrease with maturity. Instead, there is a slight increase since the accumulation of soluble cell contents (sugars) offsets the decline in cell wall digestibility. The ability to maintain high digestibility with increasing maturity gives an important advantage to sugarcane as a feed crop, especially in the critical dry season when all other grasses and forages decline in quality and availability (Preston, 1977). Furthermore, the pith also presented higher gas production (24 h) and IVDMD than the rind (Tables 1 and 4; Figure 1D). Another interesting report in this study was the similarity between the rind and pith regarding metabolizable (ME) and net energy (NE). This result is because of the contents of CP and CF in the rind. Even though the rind has a high content of NDICP, the equations for estimating ME and NE do not consider this fraction, only the CP content. Moreover, the equation does not consider the pdNDF levels. The rind, for example, showed a higher content of pdNDF than the pith (Figure 1C). All these factors may have influenced the ME and NE values.

In addition to nutritional characteristics, sugarcane's production potential and quality are essential for the sugarcane industry and animal nutrition. However, the fibrous fraction (indigestible) affects the best use of sugarcane by animals. Five different genotypes were evaluated in this study, and no differences in productivity (TSH) were observed, although G1 produced 15.36 % less than G2 (Table 5). However, the Brix and POL contents varied between genotypes (Table 5). For Barnes (1974), the higher the Brix degree, the better the nutritional value of sugarcane since approximately 90 % of sugarcane's dry matter consists of (soluble) carbohydrates. These carbohydrates

are divided into fibrous (NDF, mainly) and non-fibrous, represented mainly by sucrose, although this also contains starch and reducing sugars (glucose and fructose). Sucrose is the primary pathway through which the phloem transmits carbohydrates from leaves to the rest of the plant to provide carbon and energy for the growth and accumulation of reserve products (Felix et al., 2009). In sugarcane, ripening is a physiological process that involves the synthesis of sugars in the leaves, translocation of products, and sucrose storage in the stalk (Patrick et al., 2013). Polarization (POL) is an indicator of cane ripeness. The unripe cane has a high content of reducing sugars and color precursor compounds, resulting in low POL values with a dark-colored juice (Pereira et al., 2017). For Rhein et al. (2016), POL is one of the main characteristics of sugarcane quality, along with purity and TRS. In the case of purity, G4 and G5 genotypes showed 80.93 % and 86.47 %, respectively. Purity indicates the sucrose content and is related to the sugarcane's ripeness. The higher the purity, the greater the sucrose accumulation. As the sugarcane ages, purity tends to decrease and sugar's color may change, reducing its nutritional value. The goal is to obtain purity greater than 80 % (CONSECANA, 2006). However, the purity of the sugarcane juice can be influenced by mineral and vegetable impurities added to the sugarcane at harvest (Oliveira et al., 2012). Although the genotypes did not affect energy concentration (Tables 1 and 4), the gas production (24 h), ME, and NE showed high mean values in G2 for pith and low values in G4. It was also observed that the POL and purity values did not differ between G2 and G4. However, G2 presented 10.82 % more biomass and 10.15 % more TSH than G4 (Table 4), thereby demonstrating the potential for ruminant nutrition. As regards the reducing sugars (RS), G3 showed a lower value (16.36 %) than other genotypes (but not statistically different from G2), which means this genotype will convert less sucrose into glucose and fructose. The SR tends to follow the POL, increasing with ripening (Durán-Soria et al., 2020). In the present study the SR presented the same behavior as POL (Table 4). The TRS values showed the same behavior as RS (Table 5). The TRS indicates the total sugars in sugarcane, mainly sucrose and reducing sugars, and it is the most critical parameter for the industry and farmers (Costa et al., 2011).

Forage quality is an essential factor for adjusting intake, improving the efficiency of nutrient utilization, and reducing concentrate feedstuffs in the diet of ruminants (Tafaj et al., 2005). Low fiber digestibility is the main limiting factor for high-performance beef or dairy cattle-fed sugarcane-based diets (Corrêa et al., 2003). However, the digestibility of sugarcane does not decrease with maturity because the accumulation of soluble cell contents (sugars) offsets the decrease in cell wall digestibility (Preston, 1977).

Although the rind has a higher content of pdNDF than the pith, the high lignin content in the rind influences the quality of the final fibrous fractions of sugarcane and directly impacts the nutritional value. The genotypes do not differ nutritionally. However, the G2 presents higher biomass and energy yields than the others, making it more attractive in ruminant nutrition.

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Authors' Contributions

Conceptualization: Oliveira TS, Processi EF. **Data curation**: Fernandes AM, Moura RR, Oliveira TS. **Formal analysis**: Oliveira TS. **Investigation**: Moura RR, Camilo MG, Oliveira TS. **Methodology**: Moura RR, Aniceto ES, Silva IN. **Project administration**: Processi EF, Oliveira TS. **Resources**: Oliveira TS. **Supervision**: Oliveira TS. **Writing-original draft**: Moura RR, Processi EF, Oliveira TS. **Writing-review & editing**: Aniceto ES, Oliveira TS.

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