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RUMINANT NUTRITION

Effects of source and concentration of neutral detergent fiber from roughage in beef cattle diets on feed intake, ingestive behavior, and ruminal kinetics

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Abstract

The objectives of this research were to evaluate the effects of source and concentration of α-amylase-treated neutral detergent fiber (**aNDF**) from roughage on feed intake, ingestive behavior, and ruminal kinetics in beef cattle receiving high-concentrate diets. Six ruminally cannulated Nellore steers (408 ± 12 kg of body weight) were randomly assigned to a 6 × 6 Latin square design with six diets: 10% aNDF from corn silage (**10CS**); 20% aNDF from corn silage (**20CS**); or four diets containing 10% aNDF from corn silage and 10% aNDF from one of the following sources: sugarcane (**SC**), sugarcane bagasse (**SCB**), soybean hulls (**SH**), or low oil cottonseed hulls (**LOCH**). The parameters of passage and degradation kinetics were estimated based on a two-compartmental model with gamma- and exponential-distributed residence times. The nonlinear models were fitted by nonlinear least squares, and a linear mixed-effects model was fitted to all variables measured from the Latin square design that were related to intake, digestibility, digestion kinetic parameters, and residence times. Mean particle size (**MPS**) between roughage sources (CS, SCB, and SC) and coproducts (SH and LOCH) was affected ($P < 0.05$). Dry matter intake (**DMI**) was not affected ($P > 0.05$) by 20CS, SC, SH, or LOCH. Steers fed 20CS or LOCH diets had 16% and 20% greater DMI, respectively, ($P < 0.05$) than steers fed 10CS diet. Steers fed SCB consumed the least dry matter (**DM**). The SH and LOCH diets had lower MPS values (about 8.77 mm) in comparison to 20CS, SCB, and SC diets (about 13.08 mm) and, consequently, affected ($P < 0.05$) rumen content, ruminal in situ disappearance, nutrient digestibility, and solid fractional passage rate. Chewing time was affected ($P < 0.05$) by roughage sources and concentration. Lower values of distance travel inside the rumen (min/cm) were observed ($P < 0.05$) for the SCB and SC diets in comparison with any other diet. Except for SCB, there was no difference ($P > 0.05$) in rumen fill, among other treatments. Mean daily ruminal pH was not affected ($P > 0.05$) by 20CS, SCB, SC, and LOCH diets, and it ranged from 6.1 to 6.23. Total short-chain fatty acids concentration was affected ($P < 0.05$) by roughage source and concentration. Based on our results, we recommend that under Brazilian finishing diets, replacing roughage sources, except for SCB, based on aNDF concentration of the roughage in high-concentrate diets containing finely ground flint corn does not affect DMI.

Key words: dry matter intake, ingestive behavior, Nellore, neutral detergent fiber, roughage, rumen mat

Abbreviations

A/P	acetate to propionate ratio
ADF	acid detergent fiber
aNDF	α-amylase-treated neutral detergent fiber
aNDF	α-amylase-treated neutral detergent fiber
BCFA	branched-chain fatty acids
BW	body weight
CP	crude protein
DMED	DM effective degradability
DMI	dry matter intake
EE	ether extract
MDT	mean digestion time
MPS	mean particle size
MSP	mean time for substrate preparation (h)
NDFED	effective degradability of aNDF
NFC	nonfiber carbohydrates
RMRT	mean retention time of particles in the ruminoreticulum
SCFA	short-chain fatty acids
TMR	total mixed ration
TMRT	total mean retention time of particles in the gastrointestinal tract

Introduction

Roughage sources are included in high-concentrate finishing diets to help maintain rumen function, prevent digestive disorders, stimulate rumination, and maintain adequate ruminal pH (Campbell et al., 1992; Owens et al., 1998; Salinas-Chavira et al., 2013). Furthermore, both roughage concentration and sources influence ingestive behavior of cattle and diet utilization because they alter rumen retention time and overall rates of digestion and passage of the diet (Kreikemeier et al., 1990; Vieira et al., 2008a, 2008b, Weiss et al., 2017).

In a recent survey of 33 feedlot cattle nutrition consultants in Brazil, Oliveira and Millen (2014) reported that the concentration of neutral detergent fiber (**NDF**) in the diets ranged from 9% to 30% as added roughage and averaged 21.1% on a dry matter (**DM**) basis. The inclusion of roughage levels in diets is much broader in Brazilian than U.S. feedlots, which varied from 0% to 13.5% as added roughage on a DM basis, as reported by Vasconcelos and Galyean (2007).

Traditionally, there are many fiber sources from forages and coproducts available to the Brazilian feedlot industry (Oliveira and Millen, 2014). The roughage sources commonly used in feedlot diets in Brazil have different physical and chemical characteristics than those used in temperate regions. Few studies have evaluated roughage sources and amounts in beef feedlot animals receiving high-concentrate diets.

Galyean and Defoor (2003) suggested that roughage NDF in finishing feedlot diets influences dry matter intake (**DMI**) that are often noted when different roughage sources are fed at the same concentration. Furthermore, those authors mentioned that changing from less fibrous to more fibrous sources of roughage in the diet (small increases, e.g., 5% of DM or less), the animal typically eats more feed to maintain energy intake. However, reasons for altering feed intake, ingestive behavior, and ruminal kinetics due to changes in roughage source and level in high-concentrate diets of cattle are not fully understood. We hypothesized that replacing roughage sources based on NDF concentration (equivalent levels) in feedlot diets does not affect DMI or ruminal parameters. Therefore, the objectives of this research were to evaluate the effects of source and concentration of NDF from different roughages on DMI, ingestive behavior, physical nature of ruminal contents, ruminal metabolism, and ruminal kinetics in beef cattle receiving high-concentrate diets.

Material and Methods

All experimental procedures were approved by the Committee on Animal Use and Care at the University of São Paulo, "Luiz de Queiroz" College of Agriculture (ESALQ/USP; 2009-3).

Description of animals and diets

Six Nellore steers (408 ± 12 kg of body weight [**BW**]) were surgically prepared with rumen cannulas (silicone rubber, 10.2 cm i.d.; KEHL Indústria e Comércio LTDA—ME, São Carlos, Brazil) and individually housed in a tie-stall barn. Animals were randomly assigned to a 6 × 6 Latin square design with six treatments and six periods. Each period consisted of 10 d for diet adaptation and 9 d for sample collection. The chosen period of adaption is similar to the number of days adopted in similar studies in which intake, ingestive behavior, in situ digestibility, and ruminal fermentation were measured (Campbell et al., 1992; Shain et al., 1999; Daniel et al., 2013). Furthermore, Van Soest (1994) suggested that the average adaptation period to a new diet is about 7 to 14 d. In support of the chosen period of adaptation, Tedeschi and Fox (2018) reported that assuming an average fractional passage rate of 3.33%/h, 95% of the matter in the rumen should escape in about 90 h, let alone those degraded, and supposing that the growth rate of ruminal bacteria is greater than 3.33%/h, 4 d would be the minimum to initiate a ruminal microbiome change-over.

Steers were fed ad libitum with a total mixed ration (**TMR**) once daily at 0800 hours (5% orts allowed, DM basis). On day 11 to 19 of each period, feed intake was determined as the difference between the amounts of feed offered and refused. Animals were weighed in the morning at the beginning and at the end of each experimental period without feed and water restrictions. The six dietary treatments consisted of 10% α-amylase-treated neutral detergent fiber (**aNDF**) from corn silage (**10CS**), 20% aNDF from corn silage (**20CS**), and four diets containing 10% aNDF from corn silage and 10% aNDF from each of the following sources: sugarcane (**SC**), sugarcane bagasse (**SCB**), soybean hulls (**SH**), and low oil cottonseed hulls (**LOCH**) (Table 1). The mean particle size (**MPS**) of finely ground corn (flint type) was 1.2 mm, according to the method adopted by Yu et al. (1998). The corn silage produced in our study (whole plant) was harvested at a DM content of 32%.

Chemical analyses

The DM contents of corn silage, SCB, SC, SH, and LOCH were used to adjust diets each day throughout the study. We collected pooled samples of each roughage, concentrates, orts, feces, and rumen contents for each experimental period throughout the study to determine the chemical composition (Table 2). Dried samples of each ingredient within each period were ground to pass through a 1-mm screen (Wiley mill, MA-680 Marconi Ltda, Piracicaba, SP, Brazil), and subsamples were analyzed for DM in an air-forced oven at 105 °C for 24 h (AOAC, 1990), crude protein (**CP**) by the Dumas method (Wiles et al., 1998), ether extract (AOAC, 1990), ash (AOAC, 1980), NDF (nonsequential and ash-free; Van Soest et al., 1991), acid detergent fiber (**ADF**), and sulfuric acid lignin by the method 973.18 (Van Soest, 1973; Van Soest et al., 1991). The aNDF assay used sodium sulfite and heat-stable

Table 1. Composition of experimental diets (DM basis)

Ingredient	Experimental diets, %[1,2]					
	10CS	20CS	SC	SCB	SH	LOCH
Ingredients						
Corn silage	19.90	39.80	19.90	19.90	19.90	19.90
Finely ground corn[3]	76.20	56.20	63.60	64.70	63.00	57.10
Sugarcane	—	—	12.30	—	—	—
Sugarcane bagasse	—	—	—	11.30	—	—
Soybean hulls	—	—	—	—	13.30	—
Low oil cottonseed hulls	—	—	—	—	—	20.30
Limestone	0.80	0.80	0.80	0.80	0.80	0.80
Mineral premix[4]	1.00	1.00	1.00	1.00	1.00	1.00
Urea	2.10	2.20	2.40	2.30	2.00	0.85
Chemical composition						
TDN[5]	80.79	76.92	77.05	74.22	79.54	79.41
aNDF[6]	20.24	29.37	27.81	27.28	27.90	28.59
aNDF from roughage	11.04	21.08	19.40	19.81	20.62	21.9
CP	13.88	14.02	14.14	13.80	13.98	13.94

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SC, 10CS + 10% of aNDF from sugarcane; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]The experimental diets were formulated to contain 25 mg per kg of monensin.
[3]Mean particle size of finely ground corn was 1.30 mm.
[4]Mineral premix included: 1.0% trace mineral premix containing 215g Ca, 160g P, 14g Mg, 20g S, 260mg Co, 2.000mg Cu, 200mg 2.000mg Mn, 40mg Se, 7.200mg Zn.
[5]Total digestible nutrients (TDN) calculated according to Weiss et al. (1992).
[6]aNDF, α-amylase-treated neutral detergent fiber.

Table 2. Chemical composition of dietary components

Feed	Dietary component							
	DM	CP	EE	aNDF	ADF	Lignin	NFC	Ash
	%							
Corn silage	35.56	7.75	3.16	57.49	36.35	4.52	27.07	4.53
Finely ground corn	89.85	8.45	3.71	11.55	3.70	1.49	75.01	1.28
Sugarcane	31.04	3.91	1.21	43.49	30.41	5.61	47.29	4.10
SCB	50.40	2.83	0.80	74.06	62.48	13.93	0.65	3.80
Soybean hulls	94.48	10.45	1.48	69.03	55.53	3.38	15.62	3.42
Low oil cottonseed hulls	81.22	25.70	10.80	51.98	35.96	11.18	7.80	3.72

[1]10CS = 10% of aNDF from corn silage; 20CS = 20% of aNDF from corn silage; SC = 10CS + 10% of aNDF from sugarcane; SCB = 10CS + 10% of aNDF from sugarcane bagasse; SH = 10CS + 10% of aNDF from soybean hulls; LOCH = 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]The experimental diets were formulated to contain 25 mg per kg of monensin.
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[5]Total digestible nutrients (TDN) calculated according to Weiss et al. (1992).
[6]aNDF, α-amylase-treated neutral detergent fiber.

amylase source as recommended by the National Forage Testing Association (Undersander et al., 1993). Each sample received α-amylase (Sigma A3306; Sigma-Aldrich Brazil Ltda, São Paulo, SP, Brazil) and sodium sulfite separately for aNDF determination. Both aNDF and ADF were expressed, inclusive of residual ash. All samples were analyzed in triplicate.

Chewing activity and ruminal kinetics data

Chewing

On day 11 of each period, eating and rumination behaviors were monitored visually over a 24-h period. Eating and ruminating patterns were recorded every 5 min, and each activity was assumed to persist for the entire 5 min interval. The average intake for the period was used to estimate time spent eating or ruminating, per kilogram of DM. A period of rumination was defined as at least 5 min of ruminating activity followed by at least 5 min without ruminating activity. The total time spent chewing was calculated as the total time spent eating and ruminating (Maekawa et al., 2002). On the same day that chewing activity (day 11) was measured, samples of various roughage sources were collected for each period to measure the particle size. The particle size was determined using the Penn State Particle Size Separator (NASCO, Fort Atkinson, WI, USA; Lammers et al., 1996) adapted with a top sieve (>38, 19 to 38, 8 to 19, and <8 mm).

In situ digestibility

Eight sets of pre-weighed nylon bags (10 × 15 cm with 50 μm pore size) were placed in the rumen before feeding at 0800 hours on day 12. Each set consisted of triplicate bags containing dried (60 °C) and ground (5 mm) samples of each roughage source

(CS, SC, SCB, SH, and LOCH) (6 g of DM per bag, 20 mg/cm^2) (Huntington and Givens, 1995). All bags were initially positioned into the rumen ventral sac. A set of bags was removed from the rumen after 0, 6, 12, 18, 24, 36, 48, and 96 h of fermentation. The rate of in situ DM and aNDF digestion was determined for each roughage concentration per steer per period using the Generalized Compartmental Model of Digestion as described by Vieira et al. (2008a). The in situ aNDF residue was determined with sodium sulfite and heat stable amylase.

Ruminal fermentation characteristics

Samples of rumen fluid were collected from the dorsal and ventral area of the rumen every 2 h over 24 h on day 12. Rumen liquid pH was immediately determined using a handheld pH electrode (model HI8424—Hanna Instruments, São Paulo, Brazil). Approximately, 50 mL of filtered liquid was placed into bottles and stored at −20 °C. After thawing, the rumen fluid was centrifuged at 2,000 × *g* for 15 min at 4 °C to obtain the supernatant, which was used to measure the short-chain fatty acids (**SCFA**) (Palmquist and Conrad, 1971) and ammonia (Chaney and Marback, 1962) concentrations. The collection of the rumen fluid lasted no longer than 40 s per animal. Therefore, we believe that this might not have interfered with the in situ degradation.

Apparent digestibility

On day 13 of each period, diet digestibility was determined by collecting the total feces for four consecutive days. The apparent digestibility of each nutrient was calculated as the intake of a nutrient (kg/d) minus the fecal excretion of said nutrient (kg/d) divided by the intake.

Physical nature of ruminal contents

Ruminal mat consistency was measured 4 h after morning feeding on day 14 of each period as described by Welch (1982). Weights (500 g) were placed into the ventral rumen 2 h before the first measurement. After allowing the ruminal mat to stabilize for 2 h, a counterweight (1,500 g) was applied outside the rumen, and the distance that the ruminal weight ascended was recorded every second. The rate of cumulative ascension was recorded in cm/min.

Rumen fill or the pool size of aNDF in the ruminoreticulum was a function of the integrated processes of digestion and passage. Firkins et al. (1998) recognized that the Matis-Ellis age-dependent approach (Matis et al., 1989) is biologically sound and claimed for the development of an integrative approach like the first-order one, for example, as presented by Waldo et al. (1972). To estimate the attributes of the ruminoreticular digesta, one has to perform studies on in situ digestion kinetics and investigate the transit time of particulate and fluid markers. The information gathered with in situ and passage rate procedures yielded time-series profiles kinetically interpreted by using mathematical models. Afterward, the information from kinetic parameters of aNDF degradation and passage of Ytterbium (Yb)-labeled particle fiber was integrated as the result of the competing mechanisms of digestion and passage of the different fibrous sources fed to the animals.

Fiber kinetics

Digesta passage kinetics was measured using Yb as an external roughage marker on day 15. During the preparation process of the Yb-labeled roughage, the particle size of each roughage source (CS, SC, SCB, LOCH, and SH) was preserved as much as possible to those used in the experimental diet. Thus, the MPS of each roughage was similar to those listed in Table 3. The Yb-labeled particles were prepared by soaking each roughage source in a buffer acetate (0.1 M acetic acid to pH 6.0 with ammonium hydroxide) for 3 h and then overnight in the same solution with an exposure of 17 mg of Yb-acetate/kg DM. The labeled material was washed several times with distilled water and allowed to dry at 80 °C for 48 h. Each roughage sources was marked separately using the method described by de Vega and Poppi (1997). The passage rate of the rumen liquid phase was estimated using Cr-EDTA as a marker prepared by the methods of Downes and McDonald (1964). On day 15 of each period at 1100 hours, 3 h after feeding, steers were dosed through the ruminal fistula with 50 g of Yb-labeled roughage (either CS, SC, SCB, LOCH, or SH, according to of each dietary treatments) on top of the digesta mat in the anterior dorsal sac of the rumen. After dosing through the ruminal cannula, the ruminal contents were mixed by hand. At the same time, 2 liters of Cr-EDTA was administered into the rumen through the ruminal fistula. Fecal samples were collected at 0, 6, 12, 18, 24, 30, 36, 42, 48, 54, 60, 66, 72, 78, 84, 99, and 105 h after dosing. Fecal samples were analyzed for Yb and Cr-EDTA concentration, according to the method described by de Vega and Poppi (1997).

Passage rate attributes of the digesta

We used the two-pool compartmental model of digesta, that is, the GNG1 model (Matis et al., 1989) to mimic excretion profiles of fiber-bound markers in the feces of the animals fed the different roughage diets. We fitted the models to the fecal marker profiles with the NLIN procedure of SAS for each animal by a period intersection in the Latin square design. The NLIN procedure of SAS (SAS University Edition, SAS Systems Inc., Cary, NC, USA) fitted the models and the quality of fit of the different versions of the models generated by increasing the order of time dependency from zero to six we judged according to the procedures described by Vieira et al. (2008b). Vieira et al. (2008b) also described the models and parameters we used to compute residence times and rumen fill.

Table 3. The particle size distribution of different roughage sources using the Penn State Particle Separator

	Roughage sources						
Screen, mm	Corn silage	Sugarcane bagasse	Sugarcane	Soybean hulls	Low oil cottonseed hulls	SEM	P-value
	Particle, % retained on the sieve						
>36	5.30[b]	8.19[a]	5.40[b]	0.00[c]	1.30[c]	0.64	<0.001
19 to 36	15.80[b]	22.74[a]	20.36[a]	0.00[c]	19.30[ab]	1.38	<0.001
8 to 19	64.58[a]	33.12[d]	51.40[c]	20.81[d]	60.02[b]	1.36	<0.001
<8	13.10[e]	37.17[b]	22.86[d]	80.42[a]	31.50[c]	1.94	<0.001
MPS[1]	19.1[a]	19.3[a]	19.5[a]	6.44[c]	15.62[b]	0.73	<0.001

[1]MPS was determined according to Lammers et al. (1996).
[a–d]Means within a row with different superscripts differ ($P < 0.05$).

Ruminal evacuation

The complete evacuation of the ruminal content for digesta sampling was recorded on day 19, at the end of each period. During the rumen evacuation procedure, animals were not fed. The manually evacuated digesta was transferred to a container. A composite rumen digesta sample was formed from 10 individual samples collected throughout the evacuation procedure for DM determination. The rumen content weight was determined, and the digesta was placed back into the rumen. The ruminal fluid volume was calculated based on these measurements.

Parameterization of in situ kinetics

A mathematical model based on an aging process of digestion of the feed fibrous particles fitted the in situ profiles generated in the present study using the NLIN procedure of SAS. The model mimics aNDF degradation profiles, particularly those that exhibit decreasing sigmoid-shape patterns (Vieira et al., 2008a), as follows:

$$R_t = \tilde{A}\left(\delta_a^{N_a}\exp\left(-k_d t\right) + \exp\left(-\lambda_a t\right)\sum_{i=0}^{N_a-1}\left(1-\delta_a^{N_a-i}\right)\left(\lambda_a t\right)^i/i!\right) + \tilde{U} + e_t \quad (1)$$

The constant $\delta_a^{N_a}$ is the ratio $\lambda_a/\left(\lambda_a - k_d\right)R_t$ is the aNDF residue at time t (h), and $\tilde{A}$ is the potentially degradable aNDF fraction that must be prepared by hydration to solubilize microbial inhibitors, so that the effective colonization of the fibrous particle by fibrolytic microbes occurs. This sequence of events is described in mathematical terms by assuming a Gamma distribution of events over time, with parameters N_a (shape parameter as a positive integer, dimensionless) and $\lambda_a(1/h)$. Once the intimate contact between bacteria and the inner structures of the forage is established, the enzymatic breakdown of $\tilde{A}$ (g/kg) commences subsequently and degrades at a fractional first-order degradation rate $k_d(1/h)$. The asymptote $\tilde{U}$ (g/kg) is the unreactive or undegradable fraction of aNDF, and e_t is the random, homoscedastic, and independent normal error associated with each time-series data. The mean time for substrate preparation (*SPT*) for digestion is given by N_a/λ_a (h), and the mean time for substrate digestion (MDT) corresponds to $N_a/\lambda_a + 1/k_d$ (h).

Parameterization of passage kinetics

The parameters of passage kinetics belong to the age-dependent and age-independent double compartmental model described by Matis et al. (1989):

$$C_t = \begin{cases} e, \ 0 \le t \le \tau; \\ C_0 k_e\left(\delta^N \exp\left(-k_e t^*\right) - \exp\left(-\lambda_r t^*\right)\sum_{i=1}^{N}\delta^i\left(\lambda_r t^*\right)^{N-i}/\left(N-i\right)!\right) + e_t. \end{cases} \quad (2)$$

According to the assumptions described by Vieira et al. (2008b), C_t is the fecal marker concentration at time t after dosing (mg/kg), and $\delta = \lambda_r/\left(\lambda_r - k_e\right)$ and $t^* = t - \tau$ are auxiliary terms. In addition, C_0 is the initial marker concentration in the first pool or raft, and λ_r (1/h) is the asymptotic value of the age-dependent transference rate of particles from the raft to the turnover pool of particles that are eligible to escape the ruminoreticulum through the reticulo-omasal orifice at the fractional rate $k_e(1/h)$. N is the parameter of the Gamma time dependency that governs the probability of transference of a particle from the raft to the escapable pool. The transit of the marker through the omasum, abomasum, and intestines until its first detectable appearance in the feces corresponds to τ (h), and the model is under the constraint assumption $k_e < \lambda_r$.

A single-compartment model with nonexponential residence times described the Cr-EDTA fecal profiles to address the kinetics of fluid passage:

$$C_t = \begin{cases} e, \ 0 \le t \le \tau; \\ C_0\left(\lambda_l t^*\right)^{i-1}\exp\left(-\lambda_l t^*\right)/\left(\left(i-1\right)!F\right) + e_t. \end{cases} \quad (3)$$

In which, C_t, C_0t^*, and τ have the same meanings as previously described for equation (2), for a positive integer $i \ge 2$, and F as a numerical constant that varies with i from 2 to 6 as follows: 0.59635, 0.47454, 0.40857, 0.36528, and 0.33929, respectively. The expected value for the fractional passage rate, given the most suited model chosen among the five versions generated for $i = 2$ to 6, is given by $\bar{k}_l = F\lambda_l$ (Pond et al., 1988).

These models mimic the marker excretion profiles generated from the single doses of Yb and Cr-EDTA administered through the rumen cannulas to the animals in the digestion trial. The NLIN procedure of SAS fitted this model to the marker profiles. The choice of the best model to describe the marker profile for each animal relied on the minimum value of the Akaike information criterion (Akaike, 1974) corrected for small samples (Sugiura, 1978; Cavanaugh, 1997). The same procedure for model choice guided the selection of the most suited model to interpret the aNDF degradation profiles (equation 1).

Once the best model version is chosen for each marker and animal by period interaction, the following calculations were computed:

$$PMRT = N/\lambda_r + 1/k_e \quad (4)$$

$$LMRT = 1/\bar{k}_l \quad (5)$$

$$TMRT = \tau + N/\lambda_r + 1/k_e \quad (6)$$

Namely, the particles (*PMRT*, *h*) and liquid (*LMRT*, *h*) mean residence times, as well as the total mean residence times of particles in the gastrointestinal tract (*TMRT*, *h*), are computed from the parameter estimates.

Estimating rumen fill

The rumen fill or the pool size of aNDF in the ruminoreticulum was a function of the integrated processes of digestion and passage. Firkins et al. (1998) recognized that the age-dependent (Matis et al., 1989) approach is biologically sound and claimed for the development of an integrative approach like the first-order one, for example, as presented by Waldo et al. (1972). Vieira et al. (2008a, 2008b) presented such a solution as a generalized framework that can predict the effects of age-dependency and even predicts the first-order case under particular conditions. The model integrates digestion and passage to predict rumen fill as follows:

$$RF = \sum_j F_{NDF_j}\left(\tilde{A}_j\left(\sum_{i=1}^{N_j}\lambda_{r_j}^{i-1}/\left(\lambda_{r_j} + k_{d_j}\right)^i + \lambda_{r_j}^{N_j}/\left(\left(\lambda_{r_j} + k_{d_j}\right)^{N_j}\left(k_{e_j} + k_{d_j}\right)\right)\right) + \tilde{U}_j\left(N_j/\lambda_{r_j} + 1/k_{e_j}\right)\right) \quad (7)$$

The term F_{NDF_j} is the average aNDF intake (kg/d), and $\tilde{A}_j$, $\tilde{U}_j$, N_j λ_{r_j} k_{e_j}, and k_{d_j} have the same meanings and dimensions as previously described. The subscript j denotes the feed ingredient of the experimental diet.

Statistical analyses

The MIXED procedure of SAS University Edition fitted the statistical model described by equation (1):

$$y_{ijk} = \mu + \alpha_i + a_j + p_k + e_{ijk}$$

Where y_{ijk} is the dependent variable recorded for the j-th animal (a_j) receiving the i-th (α_i) diet during the k-th period (p_k) The intercept is the fixed constant term μ. Animal and period effects were random and the diet effect was set as fixed. The error term is e_{ijk}. The variance–covariance structures tested were the variance components (SIMPLE) and the autoregressive correlation (AR(1)), as suggested by Littell et al. (2006). We used the statistical model described by equation (1) to compute the least-squares means of the particle size of the roughage source and experimental diets, voluntary feed intake, ingestive behavior, ruminal content characteristics, ruminal mat consistency, ruminal fermentation characteristics, in situ kinetics, and passage kinetics, whereas rumen pH recordings were described as repeated measures over time, as follows:

$$y_{ijkl} = \mu + \alpha_i + a_j + p_k + \eta + \alpha\tau_{il} + e_{ijkl}$$

The η effect represent the repeated measurements taken over time in the rumen fluid (pH), the interaction between diet and time is $\alpha\tau_{il}$, and the error term is e_{ijkl}. A regression model was used to describe the time-series data, and the adopted significance level was 0.05 (type I error rate). To compare the diets, the adjusted Tukey test was used to avoid inflation of the type I error rate (Littell et al., 2006).

Results

Fiber particle size

The distribution of particles using the Penn State Particle Size Separator was different ($P < 0.05$) among roughage sources and experimental diets, and consequently affected by MPS ($P < 0.05$) (Tables 3 and 4). Similar MPS was observed ($P < 0.05$) across CS, SCB, and SC evaluated in our study. On the other hand, SH had the lowest value of MPS ($P < 0.05$) in comparison with all roughage sources studied. However, the MPS of LOCH was higher ($P < 0.05$) than SH (Table 3). The MPS of 20CS, SCB, and SC diet

Table 4. Particle size distribution of experimental diets with different roughage sources

Screen, mm	Experimental diets[1]						SEM	P-value
	10CS	20CS	SCB	SC	SH	LOCH		
	Particle, % retained sieve							
>36	0.00[c]	3.41[a]	2.14[ab]	2.70[ab]	1.58[b]	2.50[ab]	0.27	<0.001
19 to 36	3.53[c]	12.94[ab]	16.41[a]	7.32[bc]	2.95[c]	3.26[c]	1.29	<0.001
8 to 19	10.57[c]	37.39[a]	27.74[ab]	31.62[ab]	11.30[c]	22.17[b]	2.38	<0.001
<8	85.90[a]	48.24[b]	54.32[b]	58.35[b]	89.30[a]	81.43[a]	3.29	<0.001
MPS[2]	6.31[c]	13.45[a]	12.31[a]	13.48[a]	7.71[c]	9.83[b]	0.04	<0.001

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls. aNDF, α-amylase-treated neutral detergent fiber.
[2]MPS was determined according to method adapted from Lammers et al (1996).
[a-c]Means within a row with different superscripts differ ($P < 0.05$).

Table 5. Effects of source and level of NDF from roughage on feed intake and ingestive behavior

Item	Experimental diets[1]						SEM
	10CS	20CS	SCB	SC	SH	LOCH	
DMI, kg/d	7.93[bc]	9.20[a]	6.97[c]	8.60[ab]	8.83[ab]	9.52[a]	0.32
DMI, % of BW	1.82[bc]	2.10[a]	1.60[c]	1.94[ab]	2.00[ab]	2.20[a]	0.10
Eating, min/d	196.7	223.3	194.2	176.6	188.7	188.3	16.5
Eating, min/kg of DM	25.0	24.5	30.0	20.4	21.4	19.8	2.71
Eating, min/kg of aNDF	141.0[a]	95.2[ab]	112.1[ab]	79.7[b]	81.2[b]	76.6[b]	12.5
Ruminating, min/d	230.8[c]	407.5[ab]	453.3[a]	457.5[a]	240.0[c]	371.0[b]	20.0
Ruminating, min/kg of DM	29.2[d]	44.5[bc]	67.3[a]	53.4[b]	26.6[d]	39.1[cd]	3.00
Ruminating, min/kg of aNDF	164.7[bc]	173.0[bc]	273.4[a]	209.0[b]	102.0[d]	151.6[cd]	6.91
Chewing, min/d	427.5[c]	631.0[ab]	648.0[a]	634.1[a]	428.0[c]	576.0[b]	18.7
Chewing, min/kg of DM	54.3[cd]	69.0[bc]	96.6[a]	73.8[b]	48.4[d]	60.6[bcd]	4.63
Chewing, min/kg of aNDF	305.8[b]	268.0[b]	393.8[a]	288.4[b]	184.0[c]	235.0[bc]	19.72
Duration of first meal, min	39.2	35.0	34.4	30.0	39.7	36.6	5.1
Number of meals/d	11.8	12.0	10.0	11.3	10.1	11.8	1.2
Duration of each meal, min	19.4	19.1	18.7	15.8	18.7	16.1	1.8
Drinking water, min/d	24.2	20.3	21.1	24.1	22.2	25.0	4.1
Resting standing, min/d	329.1	253.3	234.0	222.0	254.5	249.1	41.0
Resting lying, min/d	664.5[ab]	500.0[b]	566.2[ab]	567.3[ab]	733.0[a]	594.1[ab]	46.4

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[a-d]Means within a row with different superscripts differ ($P < 0.05$).

was similar ($P > 0.05$), but the reduction of MPS was observed in 10CS and SH diets.

Dry matter intake, ingestive behavior, and ruminal content and fill

DMI (kg/d and % of BW) was not affected ($P > 0.05$) by 10CS, SC, or SH diets (Table 5). Steers fed 20CS or LOCH diets had 16% and 20% greater DMI, respectively ($P < 0.05$) than those steers fed a diet containing only 10CS. When comparing diets with 20% of aNDF of roughage, steers fed SCB had the lowest DMI.

Rumination time and chewing time (min/d, min/kg of DM, and min/kg of aNDF) were affected ($P < 0.05$) by roughage sources and concentration (Table 5). Time eating (min/d or min/kg of DM), duration of first meal per minute, number of meals per day, duration of each meal per minute, drinking water per minute per day, and resting standing per minute per day were not affected ($P > 0.05$) by roughage source and concentration (Table 5).

Ruminal contents such as rumen DM (%) and dry weight (kg) were affected ($P < 0.05$) by roughage source and concentration (Table 6). However, the wet weight of rumen content was not affected ($P > 0.05$) by any treatment. The ruminal mat consistency varied by all experimental diets. A similar time of weight ascension inside the rumen per second ($P > 0.05$) was observed for the 20CS, SCB, SC, and LOCH diets, but lower values were observed for the 10CS and SH diets. Lower values of distance travel inside the rumen (min/cm) were observed ($P < 0.05$) for the SCB and SC diet in comparison with any other diet. Higher values of weight ascend rate inside the rumen (cm/s) were observed ($P < 0.05$) in 10CS and SH diets in comparison to 20CS, SCB, SC, and LOCH diets.

Except for sugarcane bagasse, there was no difference ($P > 0.05$) on rumen fill estimation among diets containing 20% of aNDF of roughage on DM basis (20CS, SC, SH, and LOCH) and a diet containing only 10% of aNDF from corn silage. Furthermore, steers fed 10CS and 20CS diet showed similar rumen fill values (Table 6).

Ruminal pH and ruminal fermentation characteristics

No interaction ($P > 0.05$) of experimental diet and time post-feeding ($P > 0.05$) was observed for rumen pH. Rumen pH was affected by sampling time post-feeding ($P < 0.05$) and affected by roughage source and concentration ($P < 0.05$) (Figure 1). Mean daily ruminal pH that was not affected ($P > 0.05$) by 20CS, SCB, SC, and LOCH diets ranged from 6.1 to 6.23. However, cattle fed 10CS or SH diets showed a lower value of mean daily ruminal pH (Table 7). Roughage source and concentration affected ($P < 0.05$) SCFA, propionate, butyrate, and valerate concentrations, as well as acetate to propionate ratio (**A/P**) and ammonia N (mg/dL). Nevertheless, the proportion of acetate and branched-chain fatty acids (isobutyrate + isovalerate) were not affected ($P > 0.05$) by roughage source and concentration.

Ruminal in situ disappearance and total tract apparent digestibility

Based on the heterogeneity in the variance of the asymptotic rate of substrate preparation (λ_a, 1/h) and the mean time for substrate preparation (*MSP*, h) of the ruminal in situ DM and aNDF disappearance (Tables 8 and 9), only the most relevant results are presented, without descriptive statistics. This heterogeneous behavior occurred because of variation in the data, in addition to the small amount of sampling time within the initial phase of the degradability assay, which made both characterizing the degradation pattern and analyzing the data difficult. Although we observed different fractional rates of in situ DM degradation rate of *A* (*kd*), the lack of homogeneity for lambda variances, and consequently for its reciprocal value (i.e., MSP), implied a substantial variability for detecting significant effects.

Treatments did not differ in mean digestion time (h), potentially digestible DM fraction (% of DM), and indigestible DM fraction (% of DM) on ruminal in situ DM disappearance (Table 8). The 10CS, 20CS, and LOCH diets were not affected by fractional degradation rate of *A* (k_d, 1/h), but differing ($P < 0.05$) in comparison to SC and SH diets. The SCB diets presented the lower value of fractional degradation rate of *A* (1/h). In contrast, a soluble fraction (*S*) and DM effective degradability (**DMED**) and effective degradability of aNDF (**NDFED**) was affected by roughage source (CS, SC, SCB, SH and LOCH) and concentration diets (10CS and 20CS). (Tables 8 and 9). Roughage source and concentration were not affected by the mean digestion time, the fractional degradation rate of *An* on ruminal in situ disappearance of aNDF. However, feed potentially digestible

Table 6. Effects of source and level of NDF from roughage on ruminal content characteristics and ruminal mat consistency

Items	Experimental diets[1]						
	10CS	20CS	SCB	SC	SH	LOCH	SEM
Rumen fill[2]	2.00[b]	1.96[b]	4.00[a]	3.50[ab]	1.10[b]	1.90[b]	0.08
Rumen content							
Rumen DM, g/kg	155.00[c]	195.00[ab]	213.70[a]	195.80[ab]	152.80[c]	182.90[b]	04.50
Wet weight, kg	36.66	42.70	41.55	43.06	39.19	41.70	2.67
Dry weight, kg	5.75[b]	8.35[a]	8.88[a]	8.42[a]	6.01[b]	7.64[a]	0.54
Ruminal mat consistency[2]							
Time[3], s	399[d]	1,520[bc]	1,895[ab]	2,060[a]	448[d]	1,045[c]	140
Distance[4], min/cm	56.25[a]	51.66[ab]	42.45[b]	43.00[b]	54.40[a]	47.01[ab]	2.52
Ascend rate[5], cm/s	8.51[a]	2.07[b]	1.34[b]	1.33[b]	7.76[a]	2.81[b]	0.36

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]Rumen fill, grams of NDF concentration in the ruminal content/kg of BW.
[3]As described by Welch (1982).
[4]Time of weight ascension inside the rumen.
[5]Distance travel inside the rumen.
[6]Weight ascends rate inside the rumen.
[ab]Means within a row with different superscripts differ ($P < 0.05$).

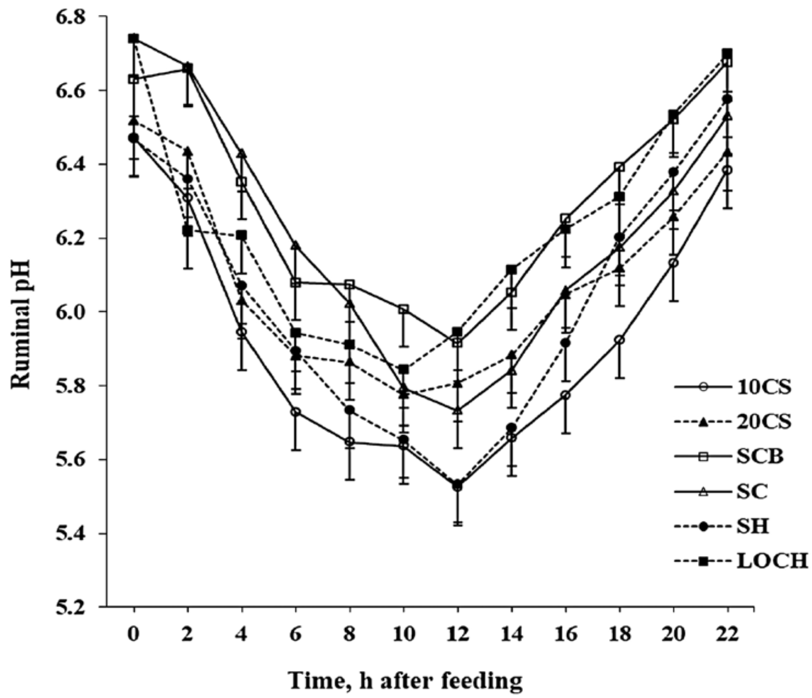

Figure 1. Rumen pH as a function of time post feeding. 10CS, 10% of NDF from corn silage; 20CS, 20% of NDF from corn silage; SCB, 10CS + 10% of NDF from sugarcane bagasse; SC, 10CS + 10% of NDF from sugarcane; SH, 10CS + 10% of NDF from soybean hulls; LOCH, 10CS + 10% of NDF from low oil cottonseed hulls. Steers were fed at 0800. No interaction of diet and time post feeding (P > 0.05) was observed..

Table 7. Effects of source and level of NDF from roughage on mean values of ruminal pH and fermentation characteristics

	Experimental diets[1]							
Item	10CS	20CS	SCB	SC	SH	LOCH	SEM	P-value
Rumen SCFA, mM								
Acetate	63.81	67.87	65.60	64.48	70.00	62.45	3.42	0.062
Propionate	29.62[a]	26.50[ab]	23.67[b]	26.80[ab]	28.17[a]	25.80[b]	0.88	0.032
Butyrate	15.50[a]	13.80[ab]	12.80[b]	14.15[ab]	14.80[ab]	13.60[ab]	0.40	<0.001
Valerate	1.20[a]	1.10[ab]	1.00[ab]	0.91[b]	1.12[a]	1.15[a]	0.04	0.004
BCFA	4.31	5.34	5.76	4.00	5.24	4.74	0.48	0.443
A/P[2]	2.20[b]	2.63[ab]	2.88[a]	2.50[ab]	2.53[ab]	2.46[ab]	0.10	0.049
Total SCFA	114.41[ab]	114.51[ab]	108.91[ab]	110.16[ab]	119.00[a]	107.94[b]	5.00	0.039
Ammonia N, mg/dL	19.31[b]	23.80[ab]	22.12[ab]	25.30[a]	23.16[ab]	13.00[c]	0.70	<0.001
Ruminal pH	5.84[c]	6.10[ab]	6.23[a]	6.20[a]	6.00[bc]	6.21[a]	0.03	<0.001

[1]10CS,10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH,10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]A/P, Acetate to propionate ratio.
[ab]Means within a row with different superscripts differ (P < 0.05).

aNDF fraction (% of DM), an indigestible aNDF fraction (% of DM), and the effective degradability of aNDF were affected by each treatment. The 10CS, 20CS, SH, and LOCH diets had similar values of potentially digestible aNDF fraction (% of DM). However, SCB and SC diets had the lowest values of potentially digestible aNDF fraction (% of DM) in comparison to other diets (Table 9).

Fractional passage rate

The fractional rate of escape of particles from the escapable pool of Yb-labeled particle fiber, as well as mean retention time of particles in the ruminoreticulum and total mean retention time of particles in the gastrointestinal tract, was affected by roughage source and concentration (Table 10). However, there was no effect on the asymptotic fractional rate of transference of particles from the raft to the pool of escaping particles and transit time (h) on any treatment evaluated in our study. In our study, the source and concentration of aNDF from roughage do not affect any liquid phase traits of the ruminoreticulum (Table 11).

Apparent digestibility

Total tract apparent digestibility of cellulose and organic matter were not affected by roughage source or roughage concentration (Table 12). The 10CS diet had greater DM total tract apparent digestibility in comparison to diets containing 20% of aNDF from roughage. Furthermore, the apparent digestibility of aNDF decreased when the corn silage concentration increased (10CS

Table 8. Effects of source and level of NDF from roughage on ruminal in situ roughage source of DM disappearance

	Experimental diets[1]							
Item[2]	10CS	20CS	SCB	SC	SH	LOCH	SEM	P-value
MDT	27.42	31.95	29.74	33.24	33.27	41.04	4.580	0.899
A	27.1	36.0	39.0	17.0	50.0	35.3	0.034	0.062
U	37.6	30.5	51.0	28.0	31.4	26.5	0.031	0.180
λ_a	684.00	650.37	1.01	0.24	313.56	1,272.59	57.50	—
MSP	2.565	0.002	16.471	14.447	5.121	0.206	2.065	—
k_d	5.10[a]	6.40[a]	1.00[c]	2.50[b]	3.00[b]	6.05[a]	0.740	<0.001
S	35.32[b]	33.61[b]	10.50[d]	55.14[a]	20.00[c]	38.16[b]	0.005	<0.001
DMED, %	50.88[b]	56.40[b]	22.46[c]	68.20[a]	50.00[b]	54.71[b]	0.010	<0.001

[1]10CS,10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]*A*, potentially digestible DM fraction (% of DM); U, indigestible DM fraction (% of DM); λ_a, asymptotic rate of substrate preparation (1/h); k_d, fractional degradation rate of A (1/h); *S*, soluble fraction (1-*A*-*U*); *DMED*, DM effective degradability.
[a–d]Means with different superscripts within a row differ ($P < 0.05$).

Table 9. Effects of source and level of NDF from roughage on ruminal in situ roughage source of disappearance of aNDF

	Experimental diets[1]							
Item[2]	10CS	20CS	SCB	SC	SH	LOCH	SEM	P-value
MDT	76.25	63.50	44.05	35.36	41.40	41.67	7.881	0.210
An	61.30[a]	64.10[a]	22.31[b]	37.24[b]	60.00[a]	54.00[a]	0.026	0.003
Un	38.73[b]	41.50[b]	78.45[a]	62.76[a]	39.45[b]	46.10[b]	0.034	0.008
λ_a	0.1817	196.20	0.205	0.388	293.99	779.78	80.560	—
MSP	20.344	8.643	10.655	17.917	5.930	0.002	5.011	—
k_d	3.52	3.63	8.61	9.74	6.04	3.34	0.027	0.385
NDFED	21.62[ab]	30.62[a]	15.21[b]	29.68[a]	30.61[a]	27.95[ab]	0.020	0.015

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]*An,* feed potentially digestible aNDF fraction (% of DM). *Un,* indigestible aNDF fraction (% of DM); λ_a, asymptotic rate of substrate preparation (1/h); k*d*, fractional degradation rate of An (1/h); *NDFED,* effective degradability of aNDF.
[a,b]Means with different superscripts within a row differ ($P < 0.05$).

Table 10. Effects of source and level of NDF from roughage on solid passage rate of Yb-labeled roughage source particles

	Experimental diets[1]							
Item[2]	10CS	20CS	SCB	SC	SH	LOCH	SEM	P-value
λ	0.05	1.68	0.19	0.15	0.85	0.56	0.20	0.114
k	5.00[a]	3.00[bc]	2.00[c]	4.10[abc]	4.20[ab]	5.70[a]	0.003	0.003
τ	11.07	8.78	10.55	13.31	12.83	9.90	1.723	0.775
RMRT[3]	37.0[c]	45.2[b]	67.1[a]	42.5[b]	28.9[c]	28.1[c]	2.5	<0.001
TMRT[4]	48.0[b]	54.0[b]	77.7[a]	55.8[b]	41.5b[bc]	38.0[c]	3.4	<0.001

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]λ, asymptotic fractional rate of transference of particles from the raft to the pool of escapable particles (1/h); $_k$, fractional rate of escape of particles from the escapable pool; τ, transit time (h); RMRT, mean retention time of particles in the ruminoreticulum (h); *TMRT*, total mean retention time of particles in the gastrointestinal tract (h).
[3]The variable *RMRT* was scaled to body mass as follows: $Y_{adj1} = RMRT/BM_{0.578.}$
[4]The variable *TMRT* was scaled to body mass as follows: $Y_{adj2} = TMRT/BM_{0.334.}$
[a-c]Means with different superscripts within a row differ ($P < 0.05$).

to 20CS). On the other hand, CP, ADF, and hemicellulose were not affected by corn silage concentration. CP total tract apparent digestibility decreased with diets containing 10% of aNDF from sugarcane and sugarcane bagasse in comparison to 10CS, 20CS, SH, and LOCH. The 20CS diet had a higher total tract apparent digestibility of aNDF and hemicellulose when compared with other treatments.

Discussion

DMI, ingestive behavior, and ruminal content and fill

According to the literature, roughage source and concentration can have a substantial effect on DMI of cattle fed high-concentrate diets (Defoor et al., 2002; Caetano et al., 2015; Marques et al., 2016). Intake of beef cattle fed high-concentrate,

Table 11. Effects of source and level of NDF from roughage on liquid passage rate of Cr-EDTA

	Experimental diets[1]							
Item[2]	10CS	20CS	SCB	SC	SH	LOCH	SEM	P-value
λ_L	0.096	0.102	0.106	0.101	0.109	0.135	0.004	0.108
k_L	4.61	5.00	4.30	4.30	5.00	6.00	1.117	—
τ	8.55	7.50	10.37	7.33	9.11	9.50	1.300	0.741
$RMRT_L$[3]	21.6	20.0	23.0	23.0	20.0	17.0	1.12	0.105
$TMRT_L$[4]	30.1	27.1	33.4	30.3	29.1	26.3	1.80	0.316

[1]10CS, 10% of aNDF from corn silage; 20CS, 20% of aNDF from corn silage; SCB, 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[2]λ_L, asymptotic passage rate of liquids from the first pool (1/h); k_L, fractional passage rate of liquids from the second pool (1/h); τ = transit time (h); $RMRT_L$, ruminoreticular liquid turnover (h); $TMRT_L$, total mean retention time in the gastrointestinal tract of Cr-EDTA marker (h).
[3]The variable *RMRT* was scaled to body mass as follows: $Y_{adj1} = RMRT/BM_{0.578.}$
[4]The variable *TMRT* was scaled to body mass as follows: $Y_{adj2} = TMRT/BM_{0.334.}$

Table 12. Effects of source and level of NDF from roughage on total tract apparent nutrient digestibility, g/kg

	Experimental diets[1]						
Item	10CS	20CS	SCB	SC	SH	LOCH	SEM
Total tract apparent digestibility, %							
DM	77.10[a]	68.71[b]	69.63[b]	70.22[b]	71.95[b]	70.80[b]	1.85
Crude protein	74.43[a]	68.35[a]	58.20[b]	58.46[b]	71.00[a]	68.85[a]	1.93
aNDF	60.30[bc]	66.31[a]	53.86[c]	52.35[c]	61.75[ab]	50.50[c]	2.51
ADF	47.42[abc]	52.63[ab]	45.40[c]	45.81[c]	54.80[a]	43.65[c]	2.35
Cellulose	49.21	56.04	49.87	49.97	56.32	48.60	2.60
Hemicellulose	66.98[ab]	82.92[a]	68.77[ab]	57.50[b]	70.02[ab]	59.07[b]	4.46
Organic matter	77.24	79.04	70.97	71.58	73.05	72.17	1.89

110CS,10% of aNDF from corn silage; 20CS,20% of aNDF from corn silage; SCB = 10CS + 10% of aNDF from sugarcane bagasse; SC, 10CS + 10% of aNDF from sugarcane; SH, 10CS + 10% of aNDF from soybean hulls; LOCH, 10CS + 10% of aNDF from low oil cottonseed hulls.
[a–c]Means within a row with different superscripts differ ($P < 0.05$).

grain-based diet is controlled by the energy demands of the animal and by metabolic factors, whereas intake of steers fed low-energy diets (often high-fiber diets) is controlled by physical factors such as ruminal fill and digesta passage (Conrad, 1966; Allen, 1996).

Traditionally, the roughage inclusion in finishing diets in North America is on average 8.3% to 9.0% (DM basis) (Vasconcelos and Galyean, 2007). In addition, corn silage and alfalfa (in the form of hay or silage) have been considered the primary sources of roughage used by the U.S. feedlots, followed by cottonseed hulls (Vasconcelos and Galyean, 2007). According to Zinn and Ware (2007), roughage is not economically competitive compared with cereal grains, and the low inclusion of roughage in feedlot diets is justified primarily as "functional" feed and only secondarily for nutrient content. Conversely, the average inclusion of roughage in feed rations in Brazilian feedlot operations is about 20% (ranging from 6% to 38%, DM basis) (Pinto and Millen, 2019) or approximately twice the concentration of oughage sed by the U.S. feedlot operations. Evaluating roughage NDF concentration in Brazilian feedlots diets, Pinto and Millen (2019) reported an average of 23% (ranging from 20% to 35%, DM basis) with a variety of roughage sources being used, such as corn silage, sugarcane bagasse, grass silage, sorghum silage, fresh-chopped sugarcane, whole cottonseed, soybean hulls, and low oil cottonseed hulls.

The ideal roughage concentration and roughage source in feedlot diets for maximum performance and maintaining rumen health varied with grain processing methods (Owens et al., 1997; Turgeon et al., 2010). Thus, it is essential to point out that most trials testing the roughage source and concentration in North America were developed in the great majority with yellow dent corn (soft endosperm type) using low roughage concentration in the diet (Deefor et al., 2002; Galyean and Defoor, 2003; Turgeon et al., 2010). Furthermore, the grain processing method most commonly used in the feedlots in this region is steam-flaking, followed by dry-rolling and high-moisture harvesting and storage (Vasconcelos and Galyean, 2007). In contrast, the grain used in feedlot diets in Brazil is characterized as a flint type (Pinto and Millen, 2019). Flint corn is defined by a lower starch digestibility due to its highly vitreous endosperm (compact and dense protein matrix) than soft endosperm hybrids (Correa et al., 2002), which leads to poorer animal performance and a potentially increased cost of gain. The finely ground corn is the grain processing method most commonly used in Brazilian feedlots, followed by coarsely ground corn (Pinto and Millen, 2019). An investigation into roughage sources and concentrations in Nellore cattle fed flint corn processed by different methods is needed to improve animal performance and maintain rumen health in feedlot cattle diets. Caetano et al. (2015) evaluated the effects of the flint corn processing method (high-moisture corn or finely ground dry corn) with one of four concentrations of NDF of roughage (3%, 8%, 13%, and 18%, DM basis) offered to Nellore bulls. In this study, the authors reported that the roughage NDF concentration for maximum DMI differed between finely ground corn and high-moisture corn, and a higher roughage inclusion was necessary for the high-moisture corn diet. Furthermore, Caetano et al. (2015) reported a reduction of DMI with diets containing 18% NDF of roughage (27% of sugarcane silage).

In this study, except for SCB and 10CS diets, DMI was not affected by 20CS, SC, SH, and LOCH diets. Even though no difference has been detected in DMI in kg/d or % BW in cattle fed diets with the same level of roughage source (20CS, SC, SH, and LOCH), a slight difference in ingestive behavior, rumen content, total SCFA concentration, and ruminal kinetics was observed, particularly across roughage sources evaluated in our study. In agreement with our results, Galyean and Defoor (2003) suggested that NDF from roughage sources would be an effective means of exchanging roughages in beef feedlot diets to achieve equal DMI and this behavior would be associated with differences in ruminal fermentation and digesta kinetics.

Increasing roughage concentration (from 10% to 20% of aNDF from corn silage, in a 10CS and 20CS diet, respectively) increased DMI by 16%. The results from our study confirm that compensation through increased DMI was possible, and 20% of aNDF from corn silage was not sufficient to restrict rumen fill in zebu cattle. According to Jennings et al. (2019), cattle consuming a finishing diet with increasing levels of roughage (5%, 9.96%, and 15% of corn stalk, DM basis) showed increased DMI (7.7, 8.2, and 8.1 kg/d, respectively). Galyean and Defoor (2003) reported that the effects of relatively small changes in roughage concentration on DMI might reflect the energy dilution effect and, consequently cattle will increase DMI in an attempt to maintain energy intake. Similarly, Arelovich et al. (2008) suggested that small increments in roughage concentration in beef feedlot diets would likely increase total DMI with limited negative effects on gain efficiency. Furthermore, the small increments of NDF from roughage in high-grain feedlot diets might stimulate salivary secretion, including changes in fermentation end products and ruminal kinetics, that favor greater DMI (Arelovich et al., 2008; Galyean and Hubbert, 2014). Thus, this statement is in agreement with our study that roughage levels ranging from 10% to 20% of aNDF from corn silage were associated with different ingestive behavior, rumen mat consistency, pH, and ruminal kinetics.

Even though there was no difference in DMI among steers fed 10CS and SCB diets, differences were observed in ingestive behavior and rumen kinetics among 10CS and SCB diets. We believe that the 10CS diet was controlled primarily by metabolic factors as a result of a low value of rumen fill, low MPS of the diet, low ruminal mat consistency, reduced chewing time, and low rumen pH. In addition, the 10CS diet showed a low mean retention time of particles in the ruminoreticulum (h) $RMRT^*$ and low total mean retention time of particles in the gastrointestinal tract (h) $TMRT^+$. In contrast, SCB diet was likely controlled by physical factors, such as high ruminal fill value, high MPS, high chewing time, high mean ruminal pH, lower fractional rate of escape of particles from the escapable pool (*k*), low DM and aNDF digestion coefficient, and high $RMRT^*$ and $TMRT^+$. The numerically greater rumen fill values observed in animals fed SCB and SC diets observed in our study are in agreement with Oliveira et al. (2011). These authors stated that the greatest effect of ruminal repletion of sugarcane NDF was caused by the high indigestible fraction of the fiber.

Armentano and Pereira (1997) mentioned that the use of NDF as the only measure of fiber contribution of a given feed turns out to be ineffective for two classes of feeds: coproducts and forages processed into various physical forms. Thus, our study confirmed that SH and LOCH diets had lower MPS values (average of 8.77 mm) in comparison to 20CS, SCB, and SC, with MPS near 13.08 mm. Consequently, SH and LOCH affected rumination time and ruminal mat consistency, suggesting low saliva buffer flow and altered ruminal fermentation and ruminal kinetics. However, it is important to note that the small addition of SH or LOCH (only 10% of aNDF, on DM basis) in the diet was not sufficient to alter DMI when compared with 20CS and SC diets, and the aNDF content of these coproducts was not a good predictor of the effects on ruminal kinetic.

There is a lack of knowledge related to how roughage sources influences feeding behavior in beef cattle fed a high-concentrate diet, and the results available in the literature are not clear. Welch (1982) stated that rumination period ranges from 30 s to more than 2 h, with 10 to 20 periods each day, to a maximum of about 8 to 9 h per day for forage-fed cattle. In contrast, according to Beauchemin et al. (1994), mature cattle fed all-concentrate diets ruminated 1.25 h per day when fed whole corn grain, and 2 to 2.5 h per day when fed whole wheat or a whole barley grain. In our study, the rumination time ranged from 3.83 to 7.62 h per day, with 10 to 12 periods each day. Moreover, we observed that the time spent ruminating and chewing (min/d) and mean rumen pH by Nellore steers fed 10CS and 20CS diets progressively increased as the proportion of forage in the diet increased. Gentry et al. (2016) reported that steers spent more time ruminating when consuming a diet containing 10% of short-grind corn stalk (307.16 min/d) and least with 5% long-grind corn stalk diet (288.74 min/d).

Nonetheless, although the SH diet contained 10% of aNDF more roughage on a DM basis in comparison to the 10CS diet, the amount of soybean hulls added in the diet was not effective at increasing chewing time, suggesting less salivary buffer secretion, which leads to lower minimum ruminal pH in comparison to 20CS, SCB, SC, and LOCH diets. In general, the adoption of coproducts in feedlot diets most often related to empirical substitution values. However, as the particle size of most coproducts such as soybean hulls is smaller than traditional forage sources, Grant (1997) suggested that when the percentage of NDF from forage decreases, the residual dietary forage must have sufficient particle size because most coproducts do not stimulate chewing as effectively as long forage. As mentioned previously, we observed that diets containing equal aNDF levels formulated with different roughage sources affected chewing activity and rumen pH in beef cattle. Allen (1997) noted that NDF alone are inadequate to balance diets for cattle because fiber differs in its effectiveness in stimulating chewing, due to differences in particle length. Therefore, precision information regarding the effectiveness of roughage sources and coproducts values to replace fiber sources is critical in beef cattle nutrition. Goulart et al. (2020) presented a comparison of methods to measure the effectiveness of roughage sources commonly used in feedlot diets and found that coproducts with small particle size (e.g., SH and LOCH) had lower physical effectiveness NDF when compared to traditional roughage sources.

Research conducted to evaluate the effect of roughage source and concentration on measurements of feeding behavior in feedlot cattle (e.g., the definition of meals) is scarce. Furthermore, studies involving meal size and frequency in steers fed low levels of roughage sources in feedlots are very important for avoiding metabolic disorders (González et al., 2012). In our study, feeding time (min/d and min/kg of DM), duration of first meal (min), number of meals per day, duration of each meal (mins), drinking water (min/d), and resting, standing, and lying times (min/d) were not affected ($P > 0.05$) by roughage source and concentration. We found that diets with roughage concentrations ranging from 10% to 20% of aNDF on a DM basis and containing 25 mg/kg of monensin, in addition to offering a TMR once a day at 0800 hours, were not sufficient to change meal behavior in Nellore steers.

According to Bartley (1976), chewing activity correlates positively to saliva production and, consequently, affects passage rate from the ruminoreticulum. In our study, even knowing that roughage sources affected chewing activities, wet weight of ruminal contents (kg) was not affected by roughage source and concentration, and consequently, no differences in the liquid passage rate of Cr-EDTA were observed. Similar results were found by Moore et al. (1990), who observed no difference in the liquid passage rate in cattle fed diet containing different roughage sources (chopped alfalfa hay, cottonseed hulls, or chopped wheat straw). Furthermore, Shain et al. (1999) also did not report differences in the liquid passage rate in cattle fed all-concentrate dry-rolled corn diet or diets containing dry-rolled corn balanced to provide equal NDF from alfalfa hay, wheat straw, or corn cobs.

Somewhat small ranges in NDF from roughage in the diet affect the physical structure of ruminal contents and the stratification or layering patterns of the solid phase of the rumen (Galyean and Defoor, 2003). This statement is in agreement with our results, where ruminal mat consistency was affected by the source and concentration of roughage. Moore et al. (1987) showed a trend of cattle has an increased ruminal fill when fed a diet containing 90% of concentrate based on steam-flaked sorghum added 10% of cottonseed hulls in comparison to diets containing 10% of alfalfa or wheat straw as the roughage source. We should note that the SH and 10CS diets demonstrated similar ascension rates (cm/sec) due to their lower rumen mat thickness and lower resistance to vertical displacement (weight ascends rate inside the rumen). The lower MPS values can explain this behavior from SH, lower MPS of the SH diet, and the lower mean retention time of particles in the ruminoreticulum ($RMRT_L$, h), which leads to lower chewing time and lower rumen pH for the first 12 h post-feeding.

Similarly, a lower rumen mat consistency when SH and LOCH replaced forage sources in the cattle diet was observed by many authors (Weidner and Grant, 1994; Allen and Grant, 2000). Due to the higher consistency and thickness of the rumen mat in the SCB and SC diets, lower distance travel inside the rumen was observed for these treatments. The rumen mat of steers fed SCB and SC diets were roughly 10 cm thicker than the rumen mat of steers fed 10CS, 20CS, and SH diets, consistent with the highest chewing time values of the former.

The greater rumen pH values for steers fed diets containing SCB and SC are possibly due to increased buffering capacity from increased saliva production due to the more total time spent chewing, which was observed for these treatments. The mean rumen pH in the 10CS diet was lower than a diet with 20% of aNDF from roughage (corn silage, sugarcane bagasse, sugarcane, and low oil cottonseed hulls), even though the total SCFA concentration was similar for all diets, except for the LOCH diet. Calderon-Cortes and Zinn (1996) examined the influence of dietary forage concentration (16% and 8% of sudangrass hay) and observed that increasing forage concentration increased ruminal pH and decreased ruminal molar proportions of butyrate. Shain et al. (1999), by evaluating the effect of forage source and particle size on finishing steers, showed that mean ruminal pH for steers fed large particle size straw diets was higher. Furthermore, the same authors mentioned that steers fed the small particle size straw diet were more prone to show higher pH values when compared with steers fed all-concentrate, large particle size alfalfa, or corncob diets. Sudweeks (1977) stated that cattle fed diets that increase chewing time and saliva flow have lower concentrations of SCFA due to a dilution effect and increased A/P. Our results tend to support these conclusions. Steers fed SCB and SC diets spent more time chewing and had numerically lower total ruminal SCFA concentrations and numerically higher A/P.

The SC diet showed a higher soluble fraction value (*S*) in comparison to other diets. This result suggests that the highest soluble fraction value (*S*) of the SC diet was due to the high concentration of carbohydrate soluble of SC, represented primarily by sucrose. Conversely, the SCB diet had a high indigestible aNDF fraction value (*Un*) and a low effective *NDFED*, and consequently, a high mean retention time of particles in the ruminoreticulum (h) (*RMRT**). However, the high values of potentially digestible aNDF fraction (*An*; g/kg DM) in 20CS and SH diets promoted higher effective *NDFED* for corn silage and soybean hulls. The effective degradability of aNDF in the 10CS diet was slightly decreased than that in the 20CS diet, which could be explained by the lower level of inclusion of corn silage to the diet, by the lower ruminal pH, and by the higher fractional rate of escape of particles from the escapable pool.

Similar results for the fractional degradation rate of *An* (k_d) across experimental diets might be influenced by the differences in the solid passage rate of Yb-labeled particles, particularly for the fractional rate of escape of particles from the escapable pool (*k*) in diets containing different roughage sources and concentration. According to Galyean and Defoor (2003), changes in the passage of dietary components from the rumen could be related to changes in DMI resulting from differences in roughage sources and concentration. Some authors suggested that passage rate of roughage particles may vary from 1% to 6%/h, as this rate is influenced by the intake level of the cattle, the specific gravity of the particles in the rumen content, particle density, and the size of the feed particles in the rumen (Martz and Belyea, 1986; Owens and Goetsch, 1988; Giger-Reverdin, 2000). Values for the passage rate of roughage particles reported by Owens and Goetsch (1988) are consistent with the values of *k* presented in this study. Weidner and Grant (1994) reported that forages and coproducts (e.g., soybean hulls) interacted to decrease the passage rate of the coproducts. Quite on the opposite, we found that even including 10% of aNDF from SH in a diet with 10% of aNDF from corn silage, the high value of transit time (*k*) and lower values of *RMRT* and *TMRT* were observed for SH diet. As reported previously, total tract DM apparent digestibility decrease when roughage concentration increase. In contrast, aNDF total tract apparent digestibility increase when corn silage level increases (10CS to 20CS diet).

According to Jung and Allen (1995), lignin is the key element that limits cell-wall digestibility. In our study, the chemical composition of SCB and LOCH showed high values of lignin (13.9% and 11.2 %, DM basis, respectively) in comparison to corn silage, sugarcane, and soybean hulls (4.52%, 5.61%, and 3.38%, DM basis, respectively). Thus, Nellore steers fed SCB diet had a lower coefficient of apparent DM and aNDF digestibility in comparison to the other treatments evaluated. Furthermore, the lower coefficient of apparent aNDF and ADF digestibility for cattle fed LOCH diet is following the method of Allen and Mertens (1988), who stated that the digestibility of fiber decreases as the rate of passage increases. Animals fed LOCH diet had high DMI, high values of fractional rate of escape of particles from the escapable pool (*k*), lower *RMRT* and $TMRT^+$ particles, as well as a high content of lignin and ADF of LOCH.

Lower total tract apparent digestibility of CP was observed from SCB and SC diets. Rotta et al. (2014) evaluated the influence of CS or SC additions to finishing diets on apparent digestibility and observed greater apparent digestibility values of CP in bulls fed SC diet in comparison to CS diet. Rotta et al. (2014)

mentioned that the greatest CP digestibility observed for SC diets could be explained by the greater quantity of urea supplied in these diets (34.9 g/kg of urea plus ammonium sulfate). In our study, the inclusion of urea was around 2.4% and 2.5% from SBC and SC diet (on DM basis, respectively) and the inclusion of urea of those diets not increased the total tract apparent digestibility of CP.

Conclusion

Based on these results, recommendations can be made regarding the use of roughage into Brazilian finishing diets. Except for SCB, the replacement of 10% of aNDF of CS (on DM basis) with SC, SH, or LOCH does not affect DMI. Feedlot diets containing finely ground flint corn with 20% of aNDF of CS or a partial replacement of CS (10% of aNDF) with SC or LOCH can provide a good feed option for diets in Brazilian feedlots to maintain mean rumen pH above 6.2 and consequently prevent digestive disorders—particularly in situations where feedlot management is questionable. However, diets with only 10% of aNDF from CS (DM basis) must be used with caution on Brazilian feedlots, due to decreased ruminal mat consistency, lower total chewing time, and consequently ruminal pH values closer to 5.8. The partial substitution of CS for SCB may be performed in proportions less than 10% of aNDF (DM basis), to avoid lowering DMI. Also, the inclusion of SCB in proportions smaller than 10% of aNDF in diets containing 10% of aNDF from CS will maintain adequate total chewing times (min/d) and adequate rumen function. Thus, SCB is considered a good alternative in diets containing high energy (high-concentrate finishing diets). Due to the lower MPS of SH, the optimal inclusion of this coproduct in diets containing a low concentration of CS (near 10% of aNDF, on DM basis) should be evaluated with caution. For instance, Nellore cattle fed diets with finely ground flint corn and 10% of aNDF from SH will require a higher minimum concentration of CS (more than 10% of NDF from corn silage) to promote harder-packed ruminal mat, stimulate rumination, and prevent a reduction in ruminal pH.

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Conflict of interest statement

The authors declare no real or perceived conflicts of interest.

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