

RUMINANT NUTRITION

Effects of source and concentration of neutral detergent fiber from roughage in beef cattle diets: Comparison of methods to measure the effectiveness of fiber

Rodrigo S. Goulart,^{†,1} Ricardo A.M. Vieira,^{‡,†} Joao L.P. Daniel,^{\$} Rafael C. Amaral,^{||} Vanessa P. Santos,^{||} Sergio G. Toledo Filho,^{||} Edward H. Cabezas-Garcia,[¶] Luis O. Tedeschi^{**}, and Luiz Gustavo Nussio^{||}

[†]Department of Animal Science, College of Animal Science and Food Engineering, University of São Paulo, Pirassununga, São Paulo 13635900, Brazil, [‡]Laboratório de Zootecnia, Universidade Estadual do Norte Fluminense, Campos dos Goytacazes, RJ, CEP 28013-602, Brazil, ^{||}Department of Animal Science, University of São Paulo, Luiz de Queiroz College of Agriculture, Piracicaba, São Paulo, 13418-900, Brazil, ^{\$}Department of Animal Science, State University of Maringá, Maringá 87020-900, Brazil, [¶]Department of Agricultural Research for Northern Sweden, Swedish University of Agricultural Sciences, SE-901 83 Umea, Sweden, ^{**}Department of Animal Science, Texas A&M University, College Station, TX 77843-2471

¹Corresponding author: rogoulart@usp.br

ORCID numbers: 0000-0002-4961-3866 (R. S. Goulart); 0000-0003-0569-899X (J. L.P. Daniel); 0000-0003-1883-4911 (L. O. Tedeschi).

Abstract

Methods have been developed to measure the effectiveness of many roughages, but few evaluations have been conducted with tropical feeds. The objectives of this research were to determine the effectiveness of roughage sources based on bioassay and laboratory methods and identify the biological attributes of the diets that correlate with these methods. Six ruminally cannulated Nellore steers (408 ± 12 kg of BW) were randomly assigned to a 6 × 6 Latin square design within six diets: negative control diet (NC) with aNDF as 10% from corn silage (CS); positive control diet (PC) with aNDF as 20% from CS; and four diets containing 10% aNDF from CS and 10% aNDF from each of the following sources: sugarcane (SC), sugarcane bagasse (SCB), soybean hulls (SH), or low oil cottonseed hulls (LOCH). Physical effectiveness factor (pef, related to the physical characteristics of aNDF) and effectiveness factor (ef, related to the ruminal pH) were determined based on a linear model approach that uses a bioassay method in which CS aNDF was assumed to be the standard fiber source. Laboratory methods to estimate pef of roughage sources were based on the proportion of DM of roughage retained on a 1.18-mm sieve pef(>1.18 mm) or retained on the 8.0-mm Penn State Particle Separator screen pef(>8.0 mm). The pef calculated by the bioassay method (total chewing time and ruminal mat resistance) for CS, SCB, and SC were higher values ($P < 0.05$) compared with SH and LOCH. The pef(rumen mat) of SC and SCB were higher ($P < 0.05$) than that of CS, SH, and LOCH. The pef(rumen mat) of LOCH was 61% higher than SH. The ef(rumen pH) of SC and LOCH was higher ($P < 0.05$) than CS and SH. The pef(chewing, min/d), pef(chewing, min/kg of DM), pef(rumen mat), and ef(rumen pH) positively correlated with rumination time, total chewing time, and ruminal mat resistance (values from transit time in seconds). No correlation was observed ($P > 0.05$) between pef(>8.0 mm) and rumination time, chewing time, and ruminal pH. The pef calculated using the bioassay method as well as pef (>8.0 mm) were negatively correlated with rumen pH ($P > 0.05$). The values of the effectiveness of fiber sources obtained in this research can be used as a guideline for nutritionists aiming to replace

roughage sources from tropical regions in beef cattle finishing diets. Under our conditions, the pef using the bioassay method or laboratory methods were not adequate in predicting ruminal pH.

Key words: bioassay method, laboratory methods, Nellore, physical effectiveness factor, roughage source

Abbreviations

ADF	acid detergent fiber
aNDF	amylase-treated neutral detergent fiber
CP	crude protein
ef(rumen pH)	effectiveness factor from mean rumen pH
ef	effectiveness factor
eNDF	effective NDF
MPS	mean particle size
pef(>1.18 mm)	physical effectiveness factor based on DM of forage retained on 1.18-mm sieve
pef(>8.0 mm)	physical effectiveness factor based on DM of forage retained on the 8.0-mm sieve
pef(chewing, min/d)	physical effectiveness factor from chewing time in min/d
pef(chewing, min/kg of DM)	physical effectiveness factor from chewing time in min/kg of DM
pef(rumen mat)	physical effectiveness factor from transit time in second
pef	physical effectiveness factor
peNDF	physically effective NDF
SCFA	short-chain fatty acids
TMR	total mixed ration

Introduction

Physically effective NDF (peNDF) is defined as the fraction of fiber that stimulates chewing and contributes to the floating mat of large particles in the rumen (Mertens, 1997). For beef cattle diets, some nutrition models predict ruminal pH, passage rate, and microbial protein yield based on the peNDF concentration (NASEM, 2016; Tedeschi and Fox, 2018).

Numerous roughage sources and coproducts are available for use in the beef industry worldwide. In the Brazilian beef cattle industry, for instance, there are many options for roughage and coproducts that have various physical and chemical characteristics. Coproducts are higher in fiber content and have the potential to replace the roughage in feedlot diets (NASEM, 2016; Tedeschi and Fox, 2018). However, the small particle size of coproducts (low peNDF), in some cases, does not stimulate chewing as effectively as other roughages. According to Gentry et al. (2016), long-grind corn stalk contained more peNDF than short-grind corn stalk, whereas a diet with 10% (on DM basis) of short-grind corn stalk contained more peNDF than 5% long-grind corn stalk or 5% short-grind corn stalk diets. Consequently, these authors indicated that rumination time (min/d) was the greatest for steers consuming a diet with 10% of short-grind corn stalk followed by steers consuming a diet with 5% long-grind corn stalk and was lowest for steers consuming a diet with 5% short-grind corn stalk diets.

Several studies have been conducted to investigate the effects of peNDF on feed intake, milk production, and chewing

activity in high-yielding early-lactation dairy cows (Clark and Armentano, 1993; Pereira et al., 1999; Zebeli et al., 2006). In contrast, there is little information concerning the effectiveness of roughage sources in maintaining rumen health in beef cattle fed high concentrate diets compared to traditional fiber sources, such as corn silage.

Various systems have been proposed to measure pef physical effectiveness factor (pef) in the scientific literature (Lammers et al., 1996; Armentano and Pereira, 1997; Mertens, 1997; Mooney and Allen, 1997). The pef or effectiveness factor (ef) of roughage can be determined by bioassay methods assessing the animal's response in terms of total chewing time, ruminal pH, and ruminal mat resistance (Armentano and Pereira, 1997). Furthermore, the proportion of DM retained in sieves with apertures of various diameters was proposed as a simple laboratory method that might be applicable to the routine analysis of pef (Lammers et al., 1996; Mertens, 1997). Although these concepts are related, there is a critical difference between the pef and ef. According to Armentano and Pereira (1997), the origin of ef is related to the sum of total abilities of the NDF in a feed to replace the NDF in forage or roughage in a ration so that the milk fat percentage as well as rumen pH or rumen short-chain fatty acids (SCFA) patterns are effectively maintained. On the other hand, pef is related to the physical characteristics of NDF (primarily particle size) that affect the chewing activity and the biphasic nature of ruminal contents, making this concept more restricted than ef.

There is a lack of data available in the literature regarding the effectiveness of roughage sources obtained using these methods in beef cattle. Moreover, various methods available to measure fiber particle size yield different pef values, but the results that have been obtained may not be reproducible and limit the acceptance of the peNDF concept. As of now, it is still unclear which method for measuring pef provides the most accurate estimates of chewing, ruminal pH, ruminal mat resistance, or any other animal response variable. Therefore, the objectives of this study were to determine the effectiveness of roughage sources based on bioassay and laboratory methods and to correlate these methods with biological variables.

Materials 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).

A full description of experimental procedures (excluding methods for determining the values of physical effectiveness factor and analysis) and diet composition is provided in a companion paper, which covers how the aNDF (amylase-treated neutral detergent fiber) of roughage sources and concentration in the diet affect DMI, ingestive behavior, and ruminal kinetics in feedlot cattle (Goulart et al., 2020).

Characterization of animals and diets

Six Nellore steers (408 ± 12 kg of BW) fitted with rumen cannulas (silicone rubber, 10.2 cm i.d.; Kehl® Indústria e Comércio LTDA,

São Carlos, SP, Brazil) and individually housed in a tie-stall barn were used. 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. Steers were fed a total mixed ration (TMR) ad libitum (5% orts allowed, DM basis) once a day at 0800 h. On days 11 to 19 of each period, feed intake was determined as the difference between the amounts of feed offered and refused.

We assessed the pef values of roughage (corn silage, CS; sugarcane, SC; and sugarcane bagasse, SCB) and coproduct (soybean hulls, SH; and low oil cottonseed hulls, LOCH) sources for the six experimental diets. A negative control diet (NC) consisted of aNDF as 10% from CS (50.2% aNDF on DM basis), and a positive control (PC) diet consisted of aNDF as 20% from CS. The other diets contained aNDF as 10% from CS and aNDF added as 10% of DM of SC (46.8% aNDF on DM basis), SCB (81.0% aNDF on DM basis), SH (75.1% aNDF on DM basis), or LOCH (49.2% aNDF on DM basis). We collected samples from all ingredients and diets during each period throughout the study to determine chemical composition.

Determination of pef and ef

Two measures of fiber effectiveness were used in this study based on the bioassay method. The pef was determined by animal response attributes which depend mostly on the macrophysical characteristics of particle size of the roughage sources, such as chewing time expressed in min/d pef(chewing, min/d), min/kg of DM pef(chewing, min/kg of DM), and min/kg of aNDF of roughage pef(chewing, min/kg of aNDF of roughage), and ruminal mat resistance pef(rumen mat). Conversely, the ef was determined via animal responses that integrated the physical and nonphysical characteristics of dietary carbohydrates, such as rumen pH, to define the overall effectiveness of fiber ef(rumen pH).

On day 12 of each period, pef and ef estimations from roughage and coproducts were determined using a slope ratio technique based on the bioassay method as recommended by Armentano and Pereira (1997). As reported in a previous survey conducted in Brazil (Oliveira and Millen, 2014), corn silage was the primary source of forage used in feedlot diets. Therefore, corn silage aNDF was considered to be the standard fiber source, and its pef and ef were both set to 1. Another assumption relates to the aNDF from concentrate mixtures (concentrate coefficient B_1 , from the linear model approach), which had pef and ef set to 0. The mean particle size of finely ground corn (flint type) was 1.2 mm, according to the method adapted by Yu et al. (1998). In this study, the slope ratio method considered the response of total chewing time (min/d, min/kg of DM, and min/kg of aNDF of roughage), rumen pH, and ruminal mat resistance for a chosen high (positive control) and low (negative control) roughage (corn silage in the present study) to constitute the standard response line (forage coefficient β_2 from the linear model approach). Furthermore, a third diet was defined as a test feed (coefficient β_3 from the linear model approach) and it contained aNDF as 10% DM of corn silage and aNDF as 10% DM of a test feed (SC, SCB, SH, and LOCH), which was formulated as suggested by Armentano and Pereira (1997). Responses above the standard response from test feed aNDF were used to calculate β_3 . The slope ratio technique was a simplification of this general model in which the coefficient for concentrate (β_1) was arbitrarily set to 0 and then solved for β_0 , β_2 , and β_3 . Therefore, the regression model used was $Y = \beta_0 + \beta_1 \times \text{concentrate aNDF} + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$, where Y is a response to dietary roughage, B_0 is in

units of the response variable, other coefficients are expressed in relation to aNDF in the diet, and feed aNDF is expressed as a fraction of dietary DM. Assuming $\beta_1 = 0$, the simplified model was $Y = \beta_0 + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$. Thus, the pef or ef values were computed as β_3/β_2 in our case. Physically effective NDF (peNDF) was the product of the pef and the analyzed aNDF of each roughage source evaluated in this study such that $\text{peNDF} = \text{pef} \times \text{aNDF}$.

Assuming that NDF is uniformly distributed over all particle sizes, and chewing activity is equal for all particles retained on a 1.18-mm sieve, Mertens (1997) proposed a laboratory method for estimating peNDF. Additionally, to develop a simple method for analysis of forage particle size and to characterize the particle size distribution of a sample, Lammers et al. (1996) proposed a practical separator method containing screens with 19- and 8-mm apertures and the bottom pan. According to Mertens (1997), the pef was determined as the sum of the proportions of the DM of roughage retained on the 1.18-mm sieve pef(>1.18 mm). Using the Penn State Particle Separator (PSPS) designed by Lammers et al. (1996), the pef was determined as the sum of the proportions of the DM of roughage retained on the 8.0-mm sieve pef(> 8.0 mm). Thus, the peNDF(> 1.18 mm) and peNDF(> 8.0 mm) values of each roughage were calculated by multiplying aNDF concentration of each roughage by the pef(> 1.18 mm) and pef(> 8.0 mm), respectively.

Statistical analyses

The PROC MIXED of SAS (SAS University Edition, SAS Systems Inc., Cary, NC) was used to fit the statistical model described by Eq. (1). The variance-covariance structures tested included the variance components (SIMPLE) and the autoregressive correlation (AR[1]) as suggested by Littell et al. (2006). We used the statistical model described by Eq. (1) to compute the least-squares means of the slope coefficients, pef, ef, pefNDF, and efNDF. The pef and ef were the variables that carried out most of the variation of the bioassay method. Because only primary chemical analyses were obtained, the aNDF contained only its intrinsic laboratory variations due to composite sampling. Therefore, errors associated with peNDF and effective NDF (eNDF) were not computed, only the errors associated with the slopes and slope-ratios (pef or ef). The Pearson correlation coefficients between the animal response variables and the physical effectiveness factor or effectiveness factor were used. To compare the diets, the adjusted Tukey test was used to avoid inflation of the Type I error rate (Littell et al., 2006).

$$y_{ijk} = \mu + \alpha_i + a_j + p_k + e_{ijk} \quad (1)$$

where y_{ijk} is the dependent variable recorded for the j th animal a_j , receiving the i th diet α_i , during the k th period p_k . Animal and period effects were random, and the diet effect was set as fixed. The intercept is the fixed constant term μ , and animal and period effects, as well as the error term e_{ijk} were random, and the diet effect was set as fixed.

Results

Physical effectiveness factor and effectiveness factor by the bioassay method

The slope coefficient was calculated for SC, SCB, SH, and LOCH diets, and the response of chewing was expressed in min/d, min/kg for DM, and min/kg of aNDF of roughage, rumen pH,

and ruminal mat resistance concerning the units of dietary aNDF added and compared with the NC and PC diets (Table 1). The pef(chewing, min/d) of CS, which was defined previously as a standard forage (pef = 1.00), was similar ($P > 0.05$) to the pef(chewing, min/d) values for SCB and SC (Table 2). In contrast, pef(chewing, min/d) values for SH and LOCH were lower ($P < 0.05$) than CS, SC, and SCB. However, the pef(chewing, min/kg of DM), calculated based on chewing time in minutes per kg of DM, was higher ($P < 0.05$) for SCB than for other roughage (CS and SC) and coproducts (SH and LOCH). No difference ($P > 0.05$) was observed in the pef(chewing, min/kg of DM) between SC and CS. However, the pef(chewing, min/kg

of DM) value of SC was more than double in comparison to CS when considering it as a standard feed in this study. Again, the pef(chewing, min/kg of DM) values for coproducts were lower than for roughage. The pef(chewing, min/kg of aNDF of roughage) of SC was greater ($P < 0.05$) than CS, SCB, and coproducts (SH and LOCH); however, similar results ($P > 0.05$) was observed between CS and SCB.

When the ruminal mat resistance was used as a response variable, the pef(rumen mat) of SC and SCB was higher than that of CS and coproducts (SH and LOCH) (Table 2). Nonetheless, the pef(rumen mat) of LOCH was 61% higher than that of SH. The ef(rumen pH) of SCB and LOCH was also higher ($P < 0.05$) than that of CS and SH when calculated using rumen

Table 1. Regression slopes of animal responses based on the standard diets (corn silage of low and high concentration of aNDF) as β_2 , and test feeds (low concentration of corn silage plus a test feed) as β_3 ¹

Items	Experimental diets ^{2,3}					SEM
	NC-PC	NC-SCB	NC-SC	NC-SH	NC-LOCH	
Chewing time, min/d	20.33 ^a	22.00 ^a	20.66 ^a	3.00 ^c	14.83 ^b	1.51
Chewing, min/kg DM	1.47 ^b	3.10 ^a	1.64 ^b	0.24 ^c	0.83 ^{bc}	0.28
Chewing, min/kg of aNDF of roughage	120.70 ^a	136.94 ^a	162.11 ^a	-11.02 ^b	82.23 ^{ab}	28.75
Ruminal mat resistance ⁴	112.13 ^b	149.12 ^a	166.11 ^a	6.89 ^d	64.60 ^c	12.32
Rumen pH	0.021 ^{bc}	0.043 ^a	0.030 ^{ab}	0.012 ^c	0.032 ^a	0.001

¹The regression model was used according to Armentano and Pereira (1997) based on bioassay method: $Y = \beta_0 + \beta_1 \times \text{concentrate aNDF} + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$; where Y is a response to dietary roughage, β_0 is in units of the response variable, other coefficients are expressed in relation to aNDF in the diet, and feed aNDF is expressed as a fraction of dietary DM. Assuming $\beta_1 = 0$, the simplified model was: $Y = \beta_0 + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$.

²NC, negative control, 10% of aNDF from corn silage; PC, positive control, 20% of aNDF from corn silage; SCB, NC + 10% of aNDF from sugarcane bagasse; SC, NC + 10% of aNDF from sugarcane; SH, NC + 10% of aNDF from soybean hulls; LOCH, NC + 10% of aNDF from low oil cottonseed hulls.

³NC-PC = β_2 ; NC-SCB, NC-SC, NC-SH, NC-LOCH = β_3 .

⁴Ruminal mat resistance, values from transit time in second.

^{a-d}Means within a row with different superscripts differ ($P < 0.05$).

Table 2. pef, ef, peNDF, and eNDF for different roughage sources determined by Bioassay method¹

Items	Roughage sources ²					SEM
	CS	SCB	SC	SH	LOCH	
Chewing time, min/d						
pef (chewing, min/d)	1.0 ^a	1.16 ^a	1.06 ^a	0.0 ^c	0.68 ^b	0.07
peNDF (chewing, min/d)	57.5	86.0	46.2	7.25	36.3	6.12
Chewing, min/kg DM						
pef (chewing, min/kg of DM)	1.0 ^b	2.50 ^a	1.20 ^b	0.0 ^c	0.45 ^c	0.08
peNDF (chewing, min/kg of DM)	57.5	182.9	52.3	4.48	23.3	12.84
Chewing, min/kg of aNDF of roughage						
pef (chewing, min/kg of aNDF of roughage)	1.0 ^b	1.13 ^b	1.34 ^a	-0.09 ^d	0.68 ^c	0.02
pefNDF (chewing, min/kg of aNDF)	57.5	83.7	52.2	6.2	35.3	8.12
Ruminal mat resistance						
pef (rumen mat) ³	1.0 ^b	1.35 ^a	1.50 ^a	0.0 ^d	0.61 ^c	0.09
peNDF (rumen mat) ³	57.5	100.1	65.4	3.2	31.4	6.76
Rumen pH						
ef (rumen pH) ³	1.0 ^{bc}	1.62 ^a	1.45 ^{ab}	0.66 ^c	1.66 ^a	0.16
eNDF (rumen pH) ³	57.5	120.3	63.0	48.3	86.4	10.44

¹The regression model was used according to Armentano and Pereira (1997) based on bioassay method: $Y = \beta_0 + \beta_1 \times \text{concentrate aNDF} + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$; where Y is a response to dietary roughage, β_0 is in units of the response variable, other coefficients are expressed in relation to aNDF in the diet, and feed aNDF is expressed as a fraction of dietary DM. Assuming $\beta_1 = 0$, the simplified model was: $Y = \beta_0 + \beta_2 \times \text{corn silage aNDF} + \beta_3 \times \text{test feed aNDF}$. The pef or ef values were computed as β_2 / β_3 according to the case. Physically effective NDF (peNDF) was the product of the pef and the analyzed aNDF of each roughage source evaluated in this study such that $\text{peNDF} = \text{pef} \times \text{aNDF}$.

²CS, Corn silage; SCB, Sugarcane bagasse; SC, sugarcane; SH, soybean hulls; LOCH, Low oil cottonseed hulls.

³Rumen mat, values from transit time in seconds as proposed by Welch (1982); Rumen pH, mean rumen pH value.

^{a-d}Means within a row with different superscripts differ ($P < 0.05$).

pH parameters by bioassay method (Table 2). It is essential to point out that the response of total chewing time (min/d, min/kg of DM, and min/kg of aNDF of roughage) and ruminal mat resistance used to calculate physical effectiveness factor values of SH was zero.

Physical effectiveness factor by laboratory methods

In this study, pef calculated using laboratory methods differed between roughage and coproducts (Table 3). CS displayed a higher value of pef(>1.18 mm) in comparison to other roughage sources. SC and LOCH displayed similar values of pef(>1.18 mm). The pef(>1.18 mm) of SH was higher than that of SCB. On the other hand, according to the PSPS method, the pef(>8.0 mm) values differed among all feeds evaluated in this study.

Pearson correlation coefficients between physical effectiveness factor or effectiveness factor and biological variables

The pef(chewing, min/d), pef(chewing, min/kg of DM), pef(rumen mat), and ef(rumen pH) positively correlated with rumination time, total chewing time, and ruminal mat resistance (Table 4). Nevertheless, it is essential to note that all pef calculated using the bioassay method negatively correlated with rumen pH ($P > 0.05$). However, no correlation was observed ($P > 0.05$) between pef(>8.0 mm) calculated by Lammers et al. (1996) and the response variables, rumination time in min/kg of DM, chewing time in min/kg of DM, and ruminal pH. Conversely, no correlation ($P > 0.05$) between rumination time (min/d), chewing time in min/d, and ruminal mat resistance was observed when calculating the pef(>1.18 mm) according to Mertens (1997).

Table 3. pef and peNDF for roughage sources determined by laboratory methods

	Roughage sources ¹					
Methods	CS	SCB	SC	SH	LOCH	SEM
Mertens (1997)						
pef(>1.18 mm), % ²	95.4 ^a	59.4 ^d	88.2 ^b	70.9 ^c	86.6 ^b	1.03
peNDF(>1.18 mm), % of DM ³	71.93	46.98	64.62	38.57	64.52	1.39
Lammers et al. (1996)						
pef(>8.0 mm), % ²	86.9 ^a	63.3 ^d	77.1 ^b	20.0 ^e	72.1 ^c	1.50
peNDF(>8.0 mm), % of DM ³	66.67	49.22	60.16	11.71	55.55	1.23

¹CS, Corn silage; SCB, Sugarcane bagasse; SC, sugarcane; SH, soybean hulls; LOCH, Low oil cottonseed hulls.

²pef (>1.18 mm), physical effectiveness factor based on DM of forage retained on 1.18-mm sieve as proposed by (Mertens, 1997);

pef (>8.0 mm), physical effectiveness factor based on DM of forage retained on the 8.0-mm sieve as proposed by (Lammers et al., 1996). The peNDF (>1.18 mm) and peNDF (>8.0 mm) values of each roughage were calculated by multiplying aNDF concentration of each roughage by the pef (>1.18 mm) and pef (>8.0 mm), respectively.

³g/kg of dry matter.

^{a-e}Means within a row with different superscripts differ ($P < 0.05$).

Table 4. Correlation between animal response variables and pef or ef from different methods

Items	% of DM					
	pef ¹	pef ¹	pef ¹	ef ¹	pef ²	pef ²
	chewing, min/d	chewing, min/kg of DM	rumen mat	rumen pH	>1.18 mm	>8.00 mm
Rumination						
min/d	—	0.58 ($P < 0.001$)	0.54 ($P < 0.001$)	0.47 ($P = 0.003$)	-0.14 ($P = 0.405$)	0.32 ($P = 0.054$)
min/kg of DM	0.56 ($P < 0.001$)	—	0.52 ($P < 0.001$)	0.51 ($P < 0.001$)	-0.40 ($P = 0.017$)	0.23 ($P = 0.173$)
Total chewing						
min/d	—	0.58 ($P < 0.001$)	0.50 ($P = 0.002$)	0.40 ($P = 0.020$)	-0.09 ($P = 0.586$)	0.38 ($P = 0.022$)
min/kg of DM	0.65 ($P < 0.0001$)	—	0.45 ($P = 0.006$)	0.42 ($P = 0.009$)	-0.40 ($P = 0.015$)	0.22 ($P = 0.189$)
Rumen pH	0.07 ($P = 0.669$)	0.21 ($P = 0.216$)	0.26 ($P = 0.120$)	—	-0.32 ($P = 0.052$)	0.03 ($P = 0.852$)
Ruminal mat resistance ³	0.47 ($P = 0.003$)	0.50 ($P = 0.002$)	—	0.30 ($P = 0.075$)	-0.14 ($P = 0.407$)	0.31 ($P = 0.066$)

¹pef, physical effectiveness factors as proposed by Armentano e Pereira (1997); pef (chewing, min/dia), values from chewing time in min/d; pef (chewing, min/kg of DM), values from chewing time in min/kg of DM; pef (rumen mat), values from transit time in second as proposed by Welch (1982); ef (pH ruminal), mean rumen pH values.

²pef (>1.18 mm), physical effectiveness factor based on DM of forage retained on 1.18-mm sieve as proposed by (Mertens, 1997);

pef (>8.0 mm), physical effectiveness factor based on DM of forage retained on the 8.0-mm sieve as proposed by (Lammers et al., 1996).

³Ruminal mat resistance, values from transit time in second.

Discussion

The effectiveness of fiber concept

High-concentrate diets for feedlot cattle must contain sufficient amounts of physically effective fiber to maximize feed efficiency during the feeding period (NASEM, 2016; Tedeschi and Fox, 2018). Thus, precise information regarding the effectiveness of feed at replacing roughage sources is a key aspect of beef cattle nutrition. The significant number of published articles evaluating the effectiveness of fiber resulted in the adoption of laboratory methods (Mertens, 1997; Beauchemin and Yang, 2005; Zhao et al., 2011). Conversely, data available in the literature estimated by bioassay method are limited and the number of trials based on animal response (chewing time, ruminal pH, ruminal mat resistance, milk fat) was conducted primarily, if not exclusively, with dairy cows (Clark and Armentano, 1993; Mooney and Allen, 1997; Pereira et al., 1999). Thus, data are lacking for evaluating the impact of the effectiveness of fiber in beef cattle diets.

As shown in this study, there are different methods for determining the effectiveness of NDF in a feed. Mertens (1997) proposed the concept for the development of both eNDF and peNDF. Although the concepts are related, the effectiveness of fiber in stimulating chewing activity is different from the effectiveness of fiber in maintaining rumen pH, rumen volatile fatty acids, and milk fat percentage when dairy cattle have been evaluated (Mertens, 2000). The peNDF of a feed is related primarily to particle size (physical properties of its fiber), which stimulates chewing activity and contributes to the floating mat of large particles in the rumen. As reported by Armentano and Pereira (1997), the ef and eNDF are related to the sum of total abilities of a feed to replace roughage so that the milk fat percentage as well as rumen pH or rumen SCFA patterns are effectively maintained. Therefore, the peNDF relates only to the physical characteristics of the fiber, making this more restricted than eNDF. Thus, peNDF would always be less than NDF, whereas eNDF can be less than or greater than the NDF concentration in a feed (Mertens, 2000). For this reason, in our study, the ef of each specific roughage was estimated based on rumen pH. In contrast, pef was estimated based on chewing activity (min/d, min/kg of DM and min/kg of aNDF of roughage), ruminal mat resistance, and laboratory method using a sieve pef(>1.18 mm) and pef(>8.0 mm).

We leveraged our data to explore how roughage sources can be replaced based on the effectiveness of fiber concept. In this analysis, we considered that CS aNDF was defined to be the standard fiber source, and its pef was set to 1. Thus, for example, SCB contained 74.1% of aNDF, which was 16% more effective as CS and resulted in a physically effective aNDF(chewing, min/d) content of 85.95% ($= 100 \times 0.741 \times 1.16$). Theoretically, $0.67 (= 0.577 \times 0.8595 \div 0.741)$ kg of SCB could replace 1.0 kg of CS containing 57.5% of peNDF(chewing, min/d). In contrast, 1.58 kg of LOCH is needed to replace 1.0 kg of CS because the peNDF(chewing, min/d) for LOCH was 36.3% ($= 0.577 \times 0.363 \div 0.5198$).

Physical effectiveness factor and effectiveness factor by the bioassay method

There is little information documenting the values of pef or ef in various roughage and coproducts for beef cattle diets that are calculated using different methods (bioassay and laboratory method). It is also important to correctly describe the NDF procedures used to delimit the inference space when evaluating the effectiveness of a feed (Silva et al., 2018). In our study, the

aNDF assay used sodium sulfite and a heat-stable amylase source as recommended by the National Forage Testing Association (Undersander et al., 1993). Furthermore, it is well documented that grain types and grain processing methods may not respond similarly in terms of animal performance, animal behavior, or ruminal kinetics when formulating diets containing equal quantities of forage NDF (Turgeon et al., 1983; Caetano et al., 2015; NASEM, 2016). All pef and ef values of roughage obtained in the present study were produced from diets containing finely ground flint corn (mean particle size of corn = 1.2 mm).

According to Armentano and Pereira (1997) and Welch (1982), chewing time and ruminal mat resistance are strongly related to forage content and forage particle size and it is an excellent physical response variable. However, as indicated by Sauvant et al. (1990), chewing activity is a variable that is not constant or additive for feeds in a ration. Chewing activity varies with breed, animal size, and level of intake as well as fiber concentration and particle size (Welch et al., 1970; Bae et al., 1983; Mertens, 1997). Armentano and Pereira (1997) related that variations due to animal response and experimental differences are minimized with bioassay method because pef or ef are fractions in which the animal effects in the numerator and denominator cancel out ($\text{pef} = [\text{min of chewing per kg of NDF in the test feed}] / [\text{min of chewing per kg of NDF in standard fiber}]$). Therefore, pef shows a proportional change in expected chewing response that should be relatively consistent among ruminants. Nevertheless, one limitation of the slope ratio approach in the bioassay method is that the forage is not truly standard, and estimates of effectiveness for the same test feed are trial dependent (Armentano and Pereira, 1997).

As shown in our study, the effectiveness of each roughage source differed according to each animal response variable chosen by the bioassay method (Table 2). SH and LOCH had a high concentration of aNDF on a DM basis (69.03% and 51.98%, respectively), but these coproducts were not effective enough at maintaining chewing time, ruminal mat resistance, or rumen pH as shown in the companion paper (Goulart et al., 2020). Because of the smaller particle size of SH, the pef(chewing, min/d), pef(chewing, min/kg of DM), and pef(rumen mat) in our experiment were not statistically different from 0. In contrast, when ef was based on rumen pH, LOCH displayed the same value as CS, SCB, and SC. Nonetheless, SH were 44% less effective in comparison to CS at maintaining rumen pH as determined by the bioassay method. According to Weidner and Grant (1994), replacement of 40% of the silage mixture with soybean hulls decreased ruminal mat resistance by 57% at 6-h postfeeding and reduced rumination activity 52%, and ruminal pH 6% compared with the diet without soybean hulls. In our study, the ef(rumen pH) of SCB and SC was 62% and 45% higher, respectively, when compared to that of CS. This behavior is a reflection of the results from (Goulart et al., 2020), who observed that cattle fed a diet containing 10% aNDF from SCB or SC showed higher chewing activity.

The pef(rumen mat) of SC and SCB was higher than that of CS and coproducts (SH and LOCH). According to Yang and Beauchemin (2009), increasing the intake of peNDF improves ruminal pH by increasing chewing activity and optimizing the ruminal environment, such as through the formation of a floating mat in the rumen that stimulates reticulo-ruminal contractions. In contrast, SH and LOCH produced lower values of pef(rumen mat) in comparison to other roughage sources evaluated in this study (CS, SC, and SCB). Considering that LOCH resulted in lower pef(rumen mat) in comparison to CS, this coproduct was effective at improving rumen pH (Goulart et al., 2020) and, consequently, yield 66% higher ef (rumen pH) compared to

that of animals fed CS. We believe that LOCH containing other characteristics, e.g., intrinsic buffering and soluble proteins that might have enhanced the rumen pH (Mertens, 2000), as observed by (Goulart et al., 2020).

Physical effectiveness factor by laboratory methods

Except for SCB, our study shows that the values obtained for the pef for CS, SC, SH, and LOCH were higher when estimated as pef(>1.18 mm) than when estimated as pef(>8.0 mm). The reason for this is that pef(>1.18 mm) contains a substantial number of particles retained on the screens between the 1.18- and 8.0-mm apertures.

In summary, our results are in agreement with other studies (Beauchemin et al., 2003; Kononoff et al., 2003), which reported a 30% to 50% higher peNDF of TMR when it was estimated as peNDF(>1.18 mm) compared with peNDF(>8.0 mm).

The pef (>1.18 mm) of SH was higher than that of SCB. In contrast, steers-fed SCB had 51.4% longer chewing time (min/d) compared to that of steers fed SH (648.0 versus 428.0 min/d, respectively) (Goulart et al., 2020). Based upon the PSPS, the pef (>8.0 mm) values differed among all feeds evaluated in this study. Under practical conditions, it is important to note that pef of coproducts, when estimated by laboratory methods, tends to be higher in comparison to the values from the bioassay method.

Mertens (1997) suggested that chewing activity and associated responses are good biological measures of the peNDF. However, the same author recommended that for a system to be applied, there must be a feed evaluation procedure that can be routinely used in a laboratory, or even on the farm in the case of peNDF. Several nutritional models that are currently used in the beef cattle industry require peNDF as a key input for predicting ruminal pH, passage rate, and microbial protein yield (NASEM, 2016; Tedeschi and Fox, 2018). Laboratory and on-farm measures of effectiveness fiber have become important to nutritionists, but the great variety of techniques and the lack of a standard procedure render estimates of physical effectiveness difficult, less specific, and reliable, which compromises reproducibility. Nevertheless, the measurement of particle size has some flaws. For example, Heinrichs et al. (1999) stated that the particle size of roughage before preparing the TMR differs significantly despite the same forage being used. Furthermore, those authors reported that pef values of roughage produced by laboratory methods are strongly dependent on various factors, such as the type of processed grains and variables related to processing, mixing, or delivering TMR to the cattle. Otherwise, effectiveness, as determined by the bioassay method or laboratory methods, might have limited applications because effectiveness values have not been repeatable across different types of diets (Clark and Armentano, 1993).

Pearson correlation coefficients between physical effectiveness factor or effectiveness factor and biological variables

As mentioned previously, the majority of studies evaluating the effectiveness of fiber concept were based on data from dairy cows (Clark and Armentano, 1993; Kononoff et al., 2003; Beauchemin and Yang, 2005; Yang and Beauchemin, 2006, 2007, 2009; Zebeli et al., 2012). Thus, the purpose of the correlation analysis in our study was to determine how beef cattle responses can be used to validate pef or ef.

We observed that pef values calculated using the bioassay method and pef(>8.0 mm) were not correlated ($P > 0.05$) with

rumen pH. Yang et al. (2001) reported that rumen fermentability of starch can have a larger effect on ruminal pH than on the physical characteristics of feeds. Furthermore, the dietary content of eNDF or peNDF is not sufficient to predict rumen pH. Moreover, Sarhan and Beauchemin (2015) evaluated eight empirical models for their ability to accurately predict mean ruminal pH in beef cattle fed a wide range of diets. They concluded that the ability of the current models to predict rumen pH from peNDF is limited in beef cattle, especially feedlot cattle fed high-grain diets. Additionally, the same authors stated that peNDF accounted for less than 50% of the variation in ruminal pH for beef cattle. In level 2 of the NRC (1996), peNDF was used to predict ruminal pH using the equation from Pitt et al. (1996). However, NASEM (2016) recognizes that the concept of peNDF does not account for the fermentability of feed or absorption from the rumen. Consequently, this factor is limited as a sole predictor of ruminal pH, particularly for feedlot cattle diets. Furthermore, Sarhan and Beauchemin (2015) claimed that rumen pH is affected by factors other than effectiveness of fiber, such as intake of fermentable carbohydrates, the degradation rate of carbohydrates, grain-processing effects, use of ionophores and feeding management, and current prediction models for rumen pH do not consider these factors. Similarly, Zebeli et al. (2012) published a review based on the role of physically effective fiber in high-producing dairy cattle and reported that, in many cases, peNDF was not sufficient to consistently predict rumen pH.

A negative correlation between pef(>1.18 mm) calculated by Mertens (1997) for rumination time and chewing time (min/kg of DM) and ruminal pH was observed. In contrast to our study, Yang and Beauchemin (2009) reported a positive correlation between chewing time and peNDF calculated by Lammers et al. (1996) or Kononoff et al. (2003) (using sieves with 8- and 1.18-mm apertures, respectively), which ranged from 0.55 to 0.76. These authors stated that the PSPS was a useful and practical device for determining the physical effectiveness of fiber, which was a good indication of the chewing potential for the feed for the dairy cows. In contrast to our results, mean ruminal pH was highly correlated with peNDF calculated by Lammers et al. (1996) and Kononoff et al. (2003), which was determined either for the TMR or the forage source according to Yang and Beauchemin (2009). Thus, it has been shown that the physical effectiveness of fiber provides an improved measure of different roughage sources, but this concept should be used with caution.

Conclusions

The values of the effectiveness of fiber sources obtained in this research can be used as a guideline for nutritionists aiming to replace roughage sources from tropical regions in beef cattle finishing diets. Based on the bioassay method, the effectiveness values (pef or ef) of each roughage measured were dependent on the chosen animal response variables. Thus, nutritionists should decide for a better animal response to estimate the pef or ef value to formulate optimal rations. Independent of animal response chosen according to the bioassay method, the pef or ef from sugarcane and sugarcane bagasse was higher than 1.0, suggesting that these roughage sources may yield different results, such as decreasing DMI and animal performance, when used to replace other forages such as corn silage. The primary limitation to laboratory assessment of physically effective NDF is that the method for measuring particle size has not been standardized and the estimation of pef from coproducts (e.g., soybean hulls) can be overestimated with this method. Even

knowing that pef (>8.0 mm) and pef (>1.18 mm) can be easily calculated in on-farm conditions, these methods provided weak associations between pef measured by laboratory methods with ruminal parameters for beef cattle fed high-concentrate diets. Our study supports the hypothesis that the physical effectiveness factor determined using bioassay or laboratory methods may not be the only parameter for adequately predicting ruminal pH. In order to establish requirements for effectiveness of fiber in feedlot beef cattle diets, more information is needed to determine the accuracy of all these systems to measure the effectiveness of fiber sources in diets containing different types of corn and processing methods. Furthermore, considering that the effectiveness concept does not account for the fermentability of feed or absorption of short-chain fatty acids from the rumen, other physicochemical characteristics of roughages and coproducts that influence their ability to maintain optimal ruminal function and animal health should be determined.

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Conflict of interest statement

The authors declare no real or perceived conflicts of interest.

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