

Simulation of rumen fermentation kinetics of by-products from the biodiesel industry with *in vitro* gas production technique

Simulação da cinética de fermentação ruminal de subprodutos da indústria do biodiesel através da técnica de produção de gases *in vitro*

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Abstract

The objective of this study was to investigate the rumen fermentation kinetics of 18 by-products from the biodiesel industry exhibiting potential for use in the feeding of ruminants via the *in vitro* gas production technique. The following feeds were investigated: cottonseed, *canudo de pito*, crambe, sunflower, castor seed (detoxified with lime) and soybean meals and cottonseed, peanut, babassu, crambe, palm kernel, sunflower, licuri nut, macaúba, forage radish and jatropha cakes. The evaluated parameters were total gas production (VfT), gas production from fibrous carbohydrates (VfFC), gas production from non-fibrous carbohydrates (VfNFC), the degradation rate of fibrous carbohydrates (kdFC), the degradation rate of non-fibrous carbohydrates (kdNFC) and lag time (lag). The feeds were grouped into six different groups according to rumen fermentation kinetic parameters and adopting an R² of 0.8. Forage radish cake and the meals of cottonseed, soybean, crambe and sunflower composed the first group, while the cakes of babassu and sunflower formed the second group. Canudo de pito and castor seed meals and the cakes of cottonseed, licuri and jatropha I and II formed the third group. The fourth group was composed by the cakes of crambe, palm kernel and peanut I. The fifth group was formed by peanut cake II, while macauba fruit cake formed the sixth group. The VfNFC and VfFC varied from 16.72 to 200.07 mL and from 53.09 to 242.12 mL, respectively. The mean kdFC and kdNFC values varied from 0.002 to 0.039% h⁻¹ and from 0.022 to 0.430% h⁻¹, respectively. The mean lag and VfT varied from 0.0001 to 5.2029 hours and 136.94 to 301.44 mL, respectively. A number of the products exhibited the potential to replace soybean meal, especially the forage radish cake and cottonseed, crambe and sunflower meals.

Key words: Degradation rate, fibrous carbohydrates, non-fibrous carbohydrates, ruminants

Resumo

Objetivou-se estudar a cinética de fermentação ruminal de 18 coprodutos da indústria do biodiesel, com potencial de uso na alimentação de ruminantes, através da técnica de produção de gases *in vitro*. Os alimentos estudados foram os farelos de: algodão, canudo de pito, crambe, girassol, mamona destoxificado com cal e soja e as tortas de: algodão, amendoim, babaçu, crambe, dendê, girassol, licuri, sementes de macaúba, nabo forrageiro e pinhão manso. Os parâmetros avaliados foram: produção total de gases (VFT), produção de gases proveniente dos carboidratos fibrosos (VFCF), produção de gases proveniente dos carboidratos não fibrosos (VFCNF), taxa de degradação dos carboidratos fibrosos (kdCF), taxa de

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degradação dos carboidratos não fibrosos (kdCNF) e lag time (Lag). Os alimentos foram agrupados através dos parâmetros de cinética de fermentação ruminal, utilizando-se um R^2 de 0,8, em seis diferentes grupos. A torta de nabo forrageiro e os farelos de algodão, soja, crambe e girassol compuseram o primeiro grupo, enquanto o segundo grupo foi formado pelas tortas de babaçu e de girassol. O terceiro grupo foi formado por farelo de canudo-de-pito e farelo de mamona destoxificado e pelas tortas de algodão, licuri e pinhão-manso I e II. O quarto grupo apresentou as tortas de amendoim I, crambe e dendê. O grupo cinco apresentou somente a torta de amendoim II, sendo que de forma semelhante o grupo seis foi formado apenas pela torta de macaúba. Os valores de VFCF e VFCNF variaram de 16,72 a 200,07 mL e 53,09 a 242,12 mL, respectivamente. Os valores médios de KdCF e KdCNF variaram de 0,0002 a 0,039 % h⁻¹ e 0,022 a 0,430 % h⁻¹, respectivamente. O Lag médio e o VFT variaram de 0,0001 a 5,2029 h e 136,94 a 301,44 mL. Alguns produtos possuem potencial para substituir o farelo de soja, especialmente a torta de nabo forrageiro e os farelos de algodão, crambe e girassol.

Palavras-chave: Carboidratos fibrosos, carboidratos não fibrosos, ruminantes, taxa de degradação

Introduction

The oil crisis of the past few decades, along with the demand for fuels and increasing environmental concerns, has placed liquid biofuels (such as biodiesel) in the spotlight as alternative energy sources (ABDALLA et al., 2008; SUAREZ et al., 2009).

It is estimated that by 2030, the use of biofuels in Brazil can reach 8% of the total fuel used in the transport sector (LOFRANO et al., 2013). Thus, there is a trend toward significant increases in biodiesel production and the availability of by-products, which will generate opportunities for the ruminant production system (NUNES et al., 2010). This possibility has attracted the interest of many producers. However, the utilization of these nutrients depends on basic knowledge of their chemical-nutritional composition and degradability in the rumen.

Several methods have been adopted to determine the amounts and proportions of nutrients required for optimal microbial and animal responses through predictions of nutrient availability in the rumen (MAGALHÃES et al., 2006). Among these techniques, *in vitro* digestibility assays predominate.

According to Theodorou et al. (1994), the *in vitro* gas production technique simulates the ruminal environment. Therefore, this technique makes it possible to study the kinetic parameters of the fractions constituting feeds because it reflects

the rate at which the components are fermented by rumen microorganisms and the extent of fermentation (CABRAL et al., 2002). The gas production model considers microbial growth and gas production to occur in the same proportions during consumption of the substrate, generating the by-products CO₂, methane and volatile fatty acids (SCHOFIELD et al., 1994).

Based on these observations, the objective was to evaluate the nutritional value of by-products from the biodiesel industry and to determine the best feeds for using in the ruminants diets, through the *in vitro* gas production technique.

Material and Methods

Feedstuffs

The experiments were performed at the Department of Animal Science of the Federal University of Viçosa, located in Viçosa, MG (Brazil).

It were included the following 18 samples of cakes and meals in the analysis, which are by-products of the biodiesel industry exhibiting potential for use in ruminant feeding: Meals of cottonseed (*Gossypium spp.* L.), canudo-de-pito (*Mabea fistulifera* Mart), crambe (*Crambe abyssinica*), sunflower (*Helianthus annuus*), castor seed (*Ricinus communis*) detoxified with lime and soybean (*Glycine max*). Moreover, cakes of cottonseed (*Gossypium spp.* L.), babassu

(*Orbignya speciosa*), crambe (*Crambe abyssinica*), palm kernel (*Elaeis guineensis*), sunflower (*Helianthus annuus*), licuri nut (*Syagrus coronata*), macauba fruit (*Acrocomia aculeata*), forage radish (*Raphanus sativus*), two types of jatropha (*Jatropha curcas*) and two types of peanut (*Arachis hypogaea*).

The two different samples of jatropha and peanut cakes, originating from different regions of Brazil, were included because they could potentially result in differences in the chemical-nutritional composition of the diets. Soybean meal was used as the reference (control treatment) for the other feeds.

Chemical analyses and calculations

To quantify the indigestible neutral detergent fiber (iNDF), the samples were processed in a Wiley knife mill equipped with 2 mm sieves. For the other analyses and *in vitro* incubation, a portion of each sample was ground in a 1 mm sieve.

It was quantified the dry matter (DM) (Method 930.15), mineral matter (MM) (Method 942.05) and crude protein (CP) (Method 984.13) in accordance with the methods described by AOAC (1995) (Table 1). Neutral detergent fiber (Method INCT-CA F-002/1) corrected for residual ash (Method INCT-CA M-002/1) and protein (Method INCT-CA N-004/1) (NDFap) and lignin (Method INCT-CA F-005/1) contents were quantified in accordance with the methods described by Detmann et al. (2012). To evaluate the ether extract (EE) content, the by-products were conditioned in XT4 (Ankom Technology Corp., Macedon, NY, USA) bags and subjected to extraction using the official AOCS high-temperature (AOCS, 2009) method using an XT15 extractor (Ankom Technology Corp., Macedon, NY, USA). The starch content was quantified using a modified amyloglucosidase/ α -amylase enzymatic method (McCLEARY et al., 1997).

Table 1. Average of the chemical composition of the feeds (n=3).

	DM (g kg ⁻¹)	g kg ⁻¹ of the DM							
		OM	CP	EE	NDFap	NFC	Lignin	Starch	iNDF
Cottonseed meal	876.1	919.2	541.4	21.0	232.7	124.0	27.5	18.3	188.5
Canudo de pito meal	890.2	923.0	577.5	31.5	101.3	212.7	21.1	7.0	51.1
Crambe meal	845.9	915.6	431.1	5.6	357.7	121.2	31.2	21.3	218.7
Sunflower meal	898.1	937.8	277.7	10.6	485.9	163.6	36.1	25.9	286.1
Castor seed meal	886.7	819.5	353.2	26.6	326.2	113.5	19.7	24.5	319.8
Soybean meal	889.0	931.5	492.0	10.0	150.7	278.8	11.9	145.0	10.5
Macauba fruit cake	896.5	948.5	373.1	86.6	449.1	39.7	35.1	19.3	121.3
Cottonseed cake	915.8	948.6	313.7	93.1	469.6	72.2	33.9	8.7	234.8
Peanut cake I	887.1	948.7	520.2	51.8	152.6	224.1	13.8	27.8	60.5
Peanut cake II	921.7	920.0	429.7	151.4	127.5	211.4	25.6	58.0	46.4
Babassu cake	932.0	944.5	190.6	92.0	623.0	38.9	42.2	10.3	407.1
Crambe cake	863.7	920.6	326.1	184.0	276.4	134.0	30.6	7.6	189.5
Palm kernel cake	894.1	924.4	196.0	53.7	601.0	73.6	50.4	16.1	227.5
Sunflower cake	884.1	934.1	287.8	21.1	390.0	235.2	37.4	8.5	247.5
Licuri nut cake	933.0	954.1	189.2	165.9	521.8	77.2	37.8	18.9	350.6
Forage radish cake	917.2	930.3	374.5	138.9	153.7	263.2	33.5	19.6	98.7
Jatropha cake I	919.1	932.2	253.2	133.8	422.2	122.9	40.9	9.9	347.1
Jatropha cake II	919.2	930.2	283.5	86.7	433.1	126.9	43.2	10.9	400.8

DM = dry matter; OM = organic matter; CP = crude protein; EE = ether extract; NDFap = neutral detergent fiber corrected for residual ash and protein; NFC = non-fibrous carbohydrates; iNDF = undegradable neutral detergent fibrous.

The iNDF was estimated through *in situ* incubation for 240 hours in a rumen-fistulated bull, in accordance with the procedures described by Casali et al. (2008).

To calculate non-fibrous carbohydrates (NFC), we adopted the equation proposed by Detmann and Valadares Filho (2010):

$$\text{NFC} = \text{OM} - (\text{CP} + \text{EE} + \text{NDF}_{\text{ap}})$$

where: NFC = non-fibrous carbohydrates (g kg^{-1}); OM = organic matter (g kg^{-1}); CP = crude protein (g kg^{-1}); EE = ether extract (g kg^{-1}); NDF_{ap} = neutral detergent fiber corrected for ash and protein (g kg^{-1}).

Simulation of rumen degradation

To calculate the accumulated gas production three replicates of each sample, weighing approximately 200 mg each, were examined. The samples were introduced into graduated glass syringes (HÄBERLE Labortechnik, FORTUNA® Germany) that had previously been washed and dried in an oven, in which the plunger was lubricated with liquid petroleum jelly.

During the incubation process, it was used a solution containing four parts of McDougall's buffer (McDOUGAL, 1949), adjusted to pH 6.8 (previously adjusted via CO_2 sparging for approximately 30 minutes), and one part of rumen fluid. The rumen fluid was collected from 3 rumen-fistulated crossbred steers that had been fed on a combination of corn silage and a concentrate mixture in a 60:40 forage-to-concentrate ratio. The fluid was conditioned into three thermal containers (temperature previously regulated to 39 °C) and taken to the laboratory. The rumen fluid collected from the three steers was mixed and filtered through a double layer of gauze prior to adding to the McDougall's buffer, as described above. The syringes were filled with 20 mL of the final solution and placed on an orbital shaking platform for conditioning in a temperature-controlled room at 39 °C.

Gas production readings were performed at 1, 2, 3, 4, 5, 6, 8, 10, 12, 15, 24, 27, 30, 33, 36, 48, 56, 60, 72, 96 and 120 hours after the beginning of incubation by directly analyzing the displacement of the syringe plunger.

Statistical analysis

The gas production curves were adjusted to the two-compartment model described by Schofield et al. (1994):

$$V_t = V_{f_1} \times \left\{ 1 + e^{-\left[\frac{2+4 \times \frac{\mu m_1}{V_{f_1}} \times (L-t)}{V_{f_1}} \right]} \right\}^{-1} + V_{f_2} \times \left\{ 1 + e^{-\left[\frac{2+4 \times \frac{\mu m_2}{V_{f_2}} \times (L-t)}{V_{f_2}} \right]} \right\}^{-1},$$

where: V_t = final gas volume (mL); V_{f_1} = final gas volume produced from non-fibrous carbohydrates (mL); μm_1 = maximum gas production rate of non-fibrous carbohydrates (mL h^{-1}); L = lag time (hours); t = time since the beginning of incubation (hours); V_{f_2} = final gas volume produced from fibrous carbohydrates (mL); and μm_2 = maximum gas production rate of fibrous carbohydrates (mL h^{-1}).

According to Schofield et al. (1994), the $\mu m/V_f$ ratio corresponds to the specific digestion rate (k) of the substrate, which is accepted as being equivalent to the microbial growth rate, given that the gas volume produced, microbial growth and digestion of the substrate are directly proportional.

Subsequently, the feeds were grouped according to their ruminal fermentation kinetics via Ward's minimum-variance method (WARD, 1963) using SAS software (SAS INSTITUTE INC, 2008).

Results and Discussion

Cluster analysis

The cluster analysis performed on the kinetic parameters, total gas production (V_{fT}), gas production from fibrous carbohydrates (V_{fFC}), gas production from non-fibrous carbohydrates (V_{fNFC}), the degradation rate of fibrous

carbohydrates (kdFC), the degradation rate of non-fibrous carbohydrates (kdNFC) and lag time (lag), suggested the formation of up to 17 different groups of feeds (Figure 1). However, such dispersed clustering is not interesting from a practical standpoint because it groups only feeds that exhibit an almost equal rumen degradation pattern. Therefore, we set a maximum R^2 value of 0.80 for the analysis and the feeds were grouped into six main groups, as follow: Forage radish cake and the meals of cottonseed, soybean, crambe and sunflower composed the first group, while the cakes of babassu and sunflower formed the second group. Canudo de pito and castor seed meals and the cakes of cottonseed, licuri and jatropa I and II formed the third group. The fourth group was composed by the cakes of crambe, palm kernel and peanut I. The fifth group was formed by peanut cake II, while macauba fruit cake formed the sixth group. This way, there were formed groups with a high similarity (80%) between feeds and it was avoided forming an excessive number of groups that could compromise the usability of the results.

Thus, the feeds examined in this study were clustered to increase the variability between groups and to reduce the variability within each group, thereby forming groups containing feeds that represented potential replacements for each other.

Gas production

Group I was formed by five feeds and was characterized by high VfNFC and VfFC values (Table 2). The gas volume produced from non-fibrous carbohydrates was 116.04 mL on average, showing variation within the group, ranging from 81.72 to 139.57 mL, for cottonseed meal and soybean meal, respectively. The fermentation of NFC begins immediately after incubation and displays an almost-complete and constant availability among feeds (VAN SOEST, 1967; MERTENS, 1996). Thus the variation in VfNFC observed might have been caused by the NFC content of the feeds in this

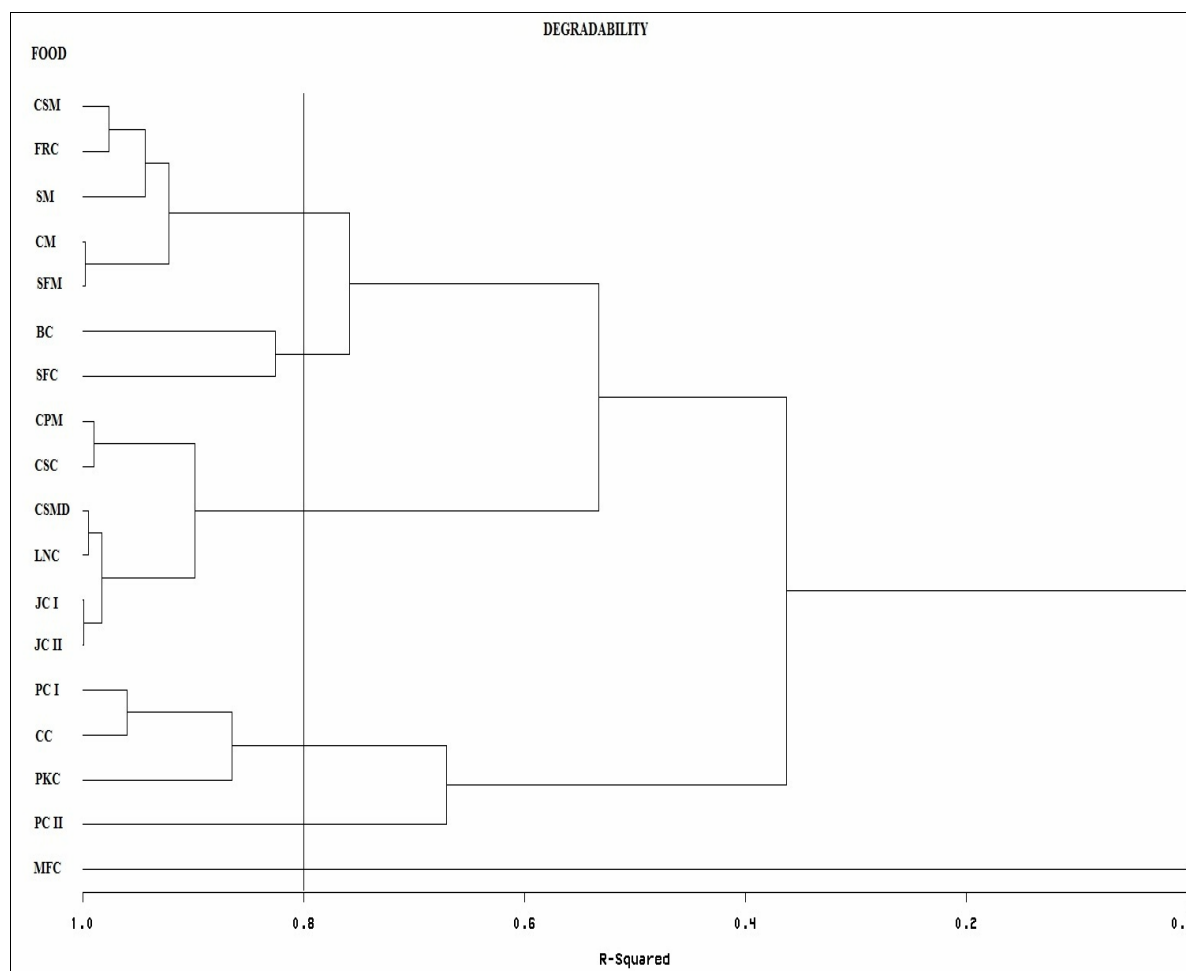
group, which ranged from 124.0 to 278.8 g kg⁻¹ (Table 1).

The average gas production from fibrous carbohydrates recorded in group I was 136.40 mL, varying from 119.00 to 161.87, for sunflower meal and soybean meal, respectively. Regarding VfFC, although soybean meal exhibited the greatest degradation of the fibrous fraction among the feeds in group I, it exhibited the lowest NDF content (150.7 g kg⁻¹). Sunflower meal, in turn, exhibited the highest NDF content among the feeds in this group (485.9 g kg⁻¹), but the degradability of FC was the lowest. This can be explained by the content of lignin that is completely non-degradable and forms a barrier that stops microorganisms from accessing potentially degradable components of the NDF fraction (FREITAS et al., 2006; MIZUBUTI et al., 2011).

The observed degradation rates ranged from 0.053 to 0.126 and 0.012 to 0.039% h⁻¹, for non-fibrous and fibrous carbohydrates, respectively. The lag values varied from 0.0011 to 1.4708 hours, for sunflower meal and forage radish cake, respectively. The EE content of forage radish cake (138.9 g kg⁻¹; the highest of the group) was possible the factor that delayed the colonization of the fibrous fraction by microorganisms, represented by the higher lag observed (1.4708 hours). It is known that elevated oil contents impair the rumen fermentation of its fiber through various mechanisms, such as physical protection of fiber and the effects of surfactants on microorganism membranes (BYERS; SCHELLING, 1988).

Group II was conspicuous by its low NFC degradation rate, with an average 41.78 mL of gas produced being produced from this fraction. The volume of gas originating from the degradation of the fibrous fraction of the feeds was 139.58 mL in this group on average. The large volume of gases produced from the degradation of NDF presented by the feeds was linked to the high average NDF content of these feeds coupled with their low NFC content, especially for babassu cake (Table 1).

Figure 1. Grouping of foods according to their degradability *in vitro*, using rumen kinetics parameters for the cluster analysis.



CSM= Cottonseed meal; FRC= forage radish cake; SM= soybean meal; CM= crambe meal; SFM= sunflower meal; BC= babassu cake; SFC= sunflower cake; CPM= canudo de pito meal; CSC= cottonseed cake; CSMD= castor seed meal detoxified; LNC= licuri nut cake; JC I= jatropha cake I; JC II= jatropha cake II; PC I= peanut cake I; CC= crambe cake; PKC= palm kernel cake; PC II= peanut cake II; MFC= macauba fruit cake.

Table 2. Kinetic parameters related to rumen degradation (n=3).

Group	Food	VfNFC ¹	kdNFC ²	Lag ³	VfFC ⁴	kdFC ⁵	VfT ⁶
Group I	Cottonseed meal	81.72	0.053	0.0048	150.34	0.012	232.06
	Forage radish cake	97.99	0.126	1.4708	130.10	0.039	228.09
	Soybean meal	139.57	0.103	1.0137	161.87	0.023	301.44
	Crambe meal	127.54	0.068	0.6611	120.68	0.020	248.22
	Sunflower meal	133.39	0.101	0.0011	119.00	0.020	252.39
	Average values	116.04	0.090	0.6303	136.39	0.0228	252.44
Group II	Babassu cake	26.83	0.298	0.0032	177.28	0.035	204.11
	Sunflower cake	56.74	0.064	2.2446	101.87	0.023	158.61
	Average values	41.78	0.181	1.1239	139.57	0.029	181.36
Group III	Canudo de pito meal	99.71	0.082	0.0323	64.89	0.016	164.60
	Cottonseed cake	110.62	0.117	0.0001	91.28	0.019	201.90
	Castor seed meal detoxified	71.37	0.072	0.0030	96.53	0.015	167.90
	Licuri nut cake	51.01	0.065	0.0025	112.38	0.002	163.39
	Jatropha cake I	59.24	0.101	0.5380	77.70	0.024	136.94
	Jatropha cake II	55.77	0.087	0.2633	85.39	0.024	141.16
	Average values	74.62	0.087	0.1398	88.03	0.016	162.65
Group IV	Peanut cake I	157.46	0.073	2.6039	91.64	0.012	249.10
	Crambe cake	126.84	0.073	1.9857	53.54	0.018	180.38
	Palm kernel cake	170.13	0.022	5.2029	53.09	0.010	223.22
	Average values	151.47	0.056	3.2641	66.09	0.013	217.56
Group V	Peanut cake II	200.07	0.109	0.0005	63.07	0.019	263.14
Group VI	Macauba fruit cake	16.72	0.430	2.8484	242.12	0.034	258.84

¹gas production from non-fibrous carbohydrates (mL); ²degradation rate of non-fibrous carbohydrates (% h⁻¹); ³lag time (h); ⁴gas production from fibrous carbohydrates (mL); ⁵degradation rate of fibrous carbohydrates (% h⁻¹); ⁶total gas production (ml).

The high NDF contents determined made it possible to use these feeds as substitutes for various roughages based on their rumen fermentation kinetics. For example, when using babassu cake as a replacement for sugarcane in the diets of heifers at rates of 0, 10, 20 and 30%, Castro (2012) did not observe alterations in dry matter intake or NDF intake. Considering the data obtained in this study, which are consistent with the data reported in the literature, we can infer that based on the observed rumen fermentation kinetics, the feeds from group II presented greater potential as replacements for roughage feeds in the ruminant diet.

The mean degradation rates for NFC and fibrous carbohydrates (FC) were 0.181 and 0.03% h⁻¹, respectively. The average lag for this group was higher than for group I, at 1.1239 hours. The degradation rates for group II were higher than for group I and the difference in lag time observed

between groups I and II was likely due to the EE content of the feeds in group II, which was higher than the majority of feeds from group I, making it difficult for the ruminal microorganisms to colonize the fibrous fraction (Table 1).

Group III was composed of six feeds and was characterized by intermediate VfNFC and VfFC values compared to the other groups. The average gas production from non-fibrous carbohydrates was 74.63 mL, varying between 51.01 and 110.62 for the licuri nut and cottonseed cakes, respectively. The gas production from fibrous carbohydrates was 88.03 mL, on average, varying between 64.89 and 112.38 mL for *canudo de pito* meal and licuri nut cake, respectively. Feeds from group III demonstrated high lignin contents, reflected in the great participation of non-degradable fiber in the NDF (Table 1). Because lignin is undegradable and reduces the degradation of the potentially

degradable fraction, it is accepted as the main factor responsible for limiting the degradation of the NDF fraction (VAN SOEST, 1994; CASSIDA et al., 2007; MIZUBUTI et al., 2011).

The degradation rates observed for fibrous and non-fibrous carbohydrates in this group were lower than in groups I and II, and the average kdNFC and kdFC values were 0.088 and 0.016% h⁻¹, respectively. Feeds with a low degradation rate, such as those in group III, are highly dependent on the passage rate for their use by ruminal microorganisms to be increased. The rumen fill effect caused by concentrate feeds is not as evident as that resulting from roughage feeds because roughage feeds exhibit a higher NDF content and physicochemical characteristics that lead to lower degradation of this fraction (OLIVEIRA et al., 2011). However, when the composition of the feeds in this group was analyzed, with the exception of *canudo de pinto* meal, we observed elevated NDFap values (Table 1), which may have caused the similar behavior of the feeds in this group compared to certain types of roughage feeds.

Considering the values corresponding to the degradation of both carbohydrate fractions and the broad variation of responses found in the literature for the feeds of group III (VIEIRA et al., 2010; SILVA et al., 2011), we can infer that these feeds should be used in animals involved in low or intermediate production levels, and that they are not good choices for diets with a high level of concentrate inclusion, in which the goal is increased escape of nutrients for digestion and absorption in the small intestine. The low degradation rates demonstrated by these feeds would result in a lower dry matter intake, possibly compromising animal performance.

Group IV exhibited high VfNFC values and intermediate VfFC values, at 151.48 and 66.09 mL, respectively. The gas production from non-fibrous carbohydrates varied from 126.84 to 170.13 mL in this group, for crambe and palm kernel cakes, respectively. However, the gas production from

fibrous carbohydrates varied from 53.09 to 91.64 mL for palm kernel cake and peanut cake I, respectively. The palm kernel cake exhibiting the highest NDF of the group IV (60.10%), and is also rich in lignin (50.4 g kg⁻¹), which might have limited the utilization of its fibrous fraction by the microorganisms. The opposite situation was observed for peanut cake I, which exhibited low lignin content (13.8 g kg⁻¹). Crambe cake demonstrated a VfFC value similar to that of palm kernel cake; however, in this case, the factor that might have limited the degradation of the fibrous fraction was the high EE content of the crambe cake compared with palm kernel (184.0 vs. 53.7 g kg⁻¹).

The average degradation rates were 0.056 and 0.13% h⁻¹ for NFC and FC, respectively. The average lag time for the group was 3.2641 hours, which was the highest value obtained among the studied groups. Palm kernel cake demonstrated high NDFap and lignin contents (60.10 and 5.04%, respectively). Therefore, palm kernel cake represents a difficult-to-degrade matrix, resulting in a low kdCF (0.010% h⁻¹). The poor degradation of the fibrous fraction coupled with the low NFC content (73.6 g kg⁻¹) may result in lower ruminal energy availability and, thus, decreased animal performance as was observed by Silva et al. (2012).

The feed that contributed most to increasing the lag time was palm kernel cake, which resulted in a lag of 5.2029 hours. Although this feed did not exhibit a high EE content (53.7 g kg⁻¹), palm kernel cake is rich in lignin (50.4 g kg⁻¹), which might have contributed to the high lag value observed.

Group V was composed of only peanut cake II and was characterized by an elevated VfNFC (200.07 mL). It was expected that Peanut cake II should demonstrate behavior similar to peanut cake I. However, the variation between these by-products, which is caused by de-standardization of the processing systems that produced the two seed samples, coupled with the dependence on the variety planted, produces differences in the

chemical-nutritional composition of the feeds and, consequently, rumen degradability (GOES et al., 2010).

The high starch content of peanut cake II (58.0 g kg⁻¹) might have positively influenced the gas production from its NFC fraction compared to the other feeds. The low V_fFC observed for this feed type, especially compared to peanut cake I, is a reflection of its lower NDF content coupled with the greater participation of lignin in the fibrous fraction (25.6 g kg⁻¹).

Group VI was also composed of a single feed type, Macauba fruit cake. And it was characterized by a lowest gas volume produced from the NFC fraction (16.72 mL), and a greatest gas volume from the fibrous fraction (242.12 mL).

Apparently, the low V_fNFC observed for Macauba fruit cake, in group VI, coupled with its high V_fFC, was caused by its low NFC content (39.7 g kg⁻¹). In addition to limiting microbial growth from this substrate, this low NFC content also stimulated increased digestion of NDF because it caused the pH to decrease. This fact, coupled with the high NDF content observed (449.1 g kg⁻¹), was likely the cause of the high V_fFC obtained for this feed type.

The lag recorded for this feed was the second highest among all of groups. This result can be explained by its high EE content (86.6 g kg⁻¹) combined with its high lignin content (35.1 g kg⁻¹), which together, can retard colonization of the fibrous fraction by the microorganisms.

Conclusions

Soybean meal displayed the best properties in relation to potential inclusion in the diets of animals. However, a number of the investigated feeds, such as the cottonseed, crambe and sunflower meals as well as forage radish cake, exhibited useful qualities. Therefore, these feeds should be further investigated using *in vivo* experiments to establish

whether they can be used in the ruminant diet and to determine the ideal levels for their inclusion.

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