

## Ingestive behavior of sheep fed Brazil nut cake in the diet

### Comportamento ingestivo de ovinos alimentados com torta de castanha-do-pará na dieta

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#### Abstract

This study evaluated the ingestive behavior of sheep when fed a corn-silage-based diet with varying levels (0%, 15%, 30%, and 60%) of Brazil nut cake (NC) (*Bertholletia excelsa* Bonpl.). Sixteen mongrel sheep with an average weight of  $33 \pm 6.04$  kg were randomly distributed between four treatments. Data were tested for assumptions of normality, subjected to an analysis of variance, and adjusted in regression equations and by Williams' test, to estimate the W point. The voluntary intake of dry matter (DM) and insoluble neutral detergent fiber (NDF) decreased linearly ( $P = 0.013$  and  $P = 0.002$ ) by 5.0 g and 2.41 g d<sup>-1</sup>, respectively, for every 1% of NC added to the diet. The time the animals spent feeding (288.75 min d<sup>-1</sup>) was not significantly affected; time spent idle increased linearly ( $P = 0.0002$ ) by 3.10 min, and time spent during rumination decreased linearly by 2.62 min ( $P = 0.001$ ) for each 1% addition of the co-product. The number of ruminated boluses (NRB) displayed a decreasing effect ( $P = 0.004$ ) of 4.61 d<sup>-1</sup> for each 1% of NC. For ruminating chews, the time spent per bolus (37.5 sec) and the number per bolus (56.14) did not differ ( $P > 0.05$ ) between treatments. However, the total chewing time (TCT) decreased linearly ( $P = 0.002$ ) by 0.05 h d<sup>-1</sup> and the number of chews per day displayed a quadratic effect ( $P = 0.008$ ), with a maximum value estimated at 17.5% of NC in the diet. Rumination efficiency did not differ between the treatments (101.95 g DM h<sup>-1</sup> and 36.76 g NDF h<sup>-1</sup>). The feeding efficiency (FE) had a linear reduction ( $P = 0.045$ ) of 0.42 g NDF h<sup>-1</sup>, but was similar for g DM h<sup>-1</sup> (172.5). The daily intake of DM and NDF showed W points estimated at 51.96% and 30.67% NC, respectively. The variables NRB, TCT, and FE (g NDF h<sup>-1</sup>) had W points estimated at 56.64%, 56.19%, and 56.33% NC, respectively. The Brazil nut cake, when present at levels greater than 56% of the diet's DM, affects the ingestive behavior of the animals, particularly rumination-related variables, and at levels of 30.67%, it begins to affect the consumption of NDF, primarily due to the ether extract content and the diet's fiber source.

**Key words:** Chewing. Co-products. Feeding. Idling period. Rumination.

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## Resumo

Avaliou-se o comportamento ingestivo de ovinos alimentados com níveis de inclusão (0, 15, 30 e 60%) de torta de castanha-do-pará (TC) (*Bertholletia excelsa* Bonpl.), na dieta à base de silagem de milho. Foram utilizados 16 cordeiros, sem raça definida e peso médio de  $33 \pm 6.04$  kg, distribuídos aleatoriamente nos quatro tratamentos. Os dados foram analisados quanto à normalidade, submetidos à análise de variância, ajustados em equações de regressão e ao teste de Williams, estimando-se o ponto W. Os consumos voluntários de matéria seca (MS) e fibra insolúvel em detergente neutro (FDN), reduziram linearmente ( $P=0,013$  e  $P=0,002$ ) em  $5,0$  g e  $2,41$  g  $\text{dia}^{-1}$ , respectivamente, para cada 1% de TC inclusa na dieta. O tempo despendido em alimentação ( $288,75$  min  $\text{dia}^{-1}$ ) não foi afetado significativamente, o tempo em ócio cresceu linearmente ( $P=0,002$ ) em  $3,10$  minutos, e o tempo gasto em ruminação decresceu ( $P=0,001$ ) em  $2,62$  minutos, para cada 1% de inclusão do coproduto. O número de bolos ruminados (NBR) sofreu efeito decrescente ( $P=0,004$ ) em  $4,61$  n°  $\text{dia}^{-1}$ . Quanto às mastigações meréricas, o tempo gasto por bolo ( $37,5$  seg) e o número por bolo ( $56,14$ ) não diferiram ( $P>0,05$ ) entre os tratamentos, mas o tempo total de mastigação (TTM) reduziu linearmente ( $P=0,002$ ) em  $0,05$  h  $\text{dia}^{-1}$  e o número de mastigações por dia apresentou efeito quadrático ( $P=0,008$ ), com máximo estimado em  $17,5\%$  de TC. A eficiência de ruminação não diferiu entre os tratamentos ( $101,95$  g MS  $\text{h}^{-1}$  e  $36,76$  g FDN  $\text{h}^{-1}$ ) e a eficiência de alimentação (EAL) sofreu redução linear ( $P=0,045$ ) em  $0,42$  g FDN  $\text{h}^{-1}$ , mas foi semelhante para g MS  $\text{h}^{-1}$  ( $172,5$ ). Os consumos diários de MS e FDN apresentaram pontos W estimados em  $51,96$  e  $30,67\%$  de TC, respectivamente. As variáveis NBR, TTM e EAL (g FDN  $\text{h}^{-1}$ ) tiveram os pontos W estimados em  $56,64$ ,  $56,19$  e  $56,33\%$  de TC. A torta de castanha-do-pará, em níveis superiores a  $56\%$  da MS da dieta, afeta o comportamento ingestivo dos animais, principalmente em variáveis relacionadas à ruminação, sendo que a partir de  $30,67\%$  já passa a afetar o consumo de FDN, tendo como principais fatores o teor de extrato etéreo e a fonte de fibra das dietas.

**Palavras-chave:** Alimentação. Castanha-do-pará. Coprodutos. Ócio. Ruminação.

## Introduction

Animal performance is highly dependent upon nutrition. Although this has been a subject of previous research, studies have not considered sheep behavior, resulting in limited interpretation of certain research findings (CARVALHO et al., 2007). The study of ingestive behavior is an important tool in the evaluation of dietary performance, as it allows the adjustment of feeding management to obtain better productive performance (CIRNE et al., 2014). Additionally, the study of ingestive behavior provides useful information for the evaluation of both well-known and unknown ingredients in livestock feed analysis. The main findings are obtained by examining behavioral parameters, such as apprehension, chewing, and rumination, relative to the partial and total time spent performing these behaviors and their efficiency (BÜRGER et al., 2000). For instance, confined animals spend around an hour consuming feeds with high energy density, whereas they spend more than six hours consuming

food sources with low energy density (HÜBNER et al., 2008). The nature of the diet also influences rumination time, with rumination time being proportional to the cell-wall content of the feed (VAN SOEST, 1994). In other words, performance and efficiency of rumination is determined by the quantity and quality of the dietary fiber in the food source.

As the development of agro-industries has expanded the production of cosmetic and food products, a consequence has been the generation and accumulation of waste. To promote a more adequate and sustainable waste management system within the industry, research is needed regarding the properties of these co-products and their applicability in animal nutrition. The Amazon region is known for its diverse plant products. Among them is the Brazil nut (*Bertholletia excelsa* Bonpl.), a product of great sociocultural importance in the northern region of Brazil, as it provides economic viability for municipalities in Pará, and is one of the best-

selling extracted products in both the domestic and export markets (SILVA et al., 2013). The extraction and processing of oil from almond seeds supports numerous communities in the Amazon and drives their regional economies while promoting forest conservation (HOMMA, 2012).

Thus, the objective of this study was to evaluate the ingestive behavior of sheep when fed a corn-silage-based diet with increasing levels (0%, 15%, 30%, and 60%) of the Brazil nut cake (NC), and to examine the following: time spent feeding, ruminating, and idling; chews per bolus, per day; total chewing time; number of ruminated boluses and time spent ruminating each; and feeding and rumination efficiency (in grams of dry matter and insoluble, neutral detergent fiber).

## Material and Methods

The experiment was conducted in an enclosed barn at the Animal Research Unit “Senator Álvaro Adolpho” in the Embrapa Eastern Amazon, in Belém, Pará (1° 28′ S and 48° 27′ W). Laboratory tests were conducted in the Animal Nutrition Laboratory at the Federal University of Pará, Castanhal Campus, and in the Animal Nutrition Laboratory at the Federal Rural University of the Amazon, Belém Campus. Sixteen mongrel sheep with an average weight of  $33 \pm 6.04$  kg were used. They were kept in individual metabolic cages, and fed a corn-silage-based diet (CS) with increasing levels (0%, 15%, 30%, and 60%) of Brazil nut cake (NC) (*Bertholletia excelsa* Bonpl.) in the dry matter (Table 1). These diets (treatments) will be referred to as “T”.

**Table 1.** Chemical composition of Brazil nut cake (NC) and diets with 0%, 15%, 30% and 60% inclusion of NC in the dry matter (DM) the corn-silage-based diet.

Component (% DM)	Inclusion level (% DM)				
	0**	15	30	60	NC
Dry matter*	33.07	40.99	48.90	64.74	85.85
Organic matter	95.21	94.35	93.49	91.77	89.48
Mineral matter	4.79	5.65	6.51	8.23	10.52
Ashes insoluble in neutral detergent	1.05	1.18	1.31	1.56	1.90
Ashes insoluble in acid detergent	0.76	0.80	0.85	0.93	1.05
Crude protein	7.88	11.95	16.03	24.17	35.03
Insoluble protein in neutral detergent	0.71	2.10	3.49	6.28	9.99
Insoluble protein in acid detergent	0.24	0.63	1.02	1.81	2.85
Ether extract	4.38	5.97	7.57	10.75	15.00
Insoluble fiber in neutral detergent <sub>ap</sub>	38.21	37.06	35.91	33.61	30.54
Insoluble fiber in acid detergent <sub>ap</sub>	19.25	18.61	17.98	16.70	15.00
Lignin	2.18	2.47	2.77	3.35	4.13
Hemicellulose	18.96	18.45	17.93	16.91	15.54
Cellulose	17.07	16.14	15.21	13.35	10.87
Non-fiber carbohydrates	44.74	39.37	33.99	23.24	8.91
Total digestible nutrients	74.38	79.51	73.61	69.01	-
Crude energy (cal gDM <sup>-1</sup> )	3435.24	3544.41	3653.57	3871.90	4163.01

\*% natural matter; \*\*corn-silage; ap = corrected for ash and crude protein; NC= Brazil nut cake.

The co-product was obtained from the Amazon Oil Industry agribusiness in Ananindeua, Pará, Brazil, where the oil is extracted by mechanically pressing the hull, containing Brazil nuts, with the

addition of rice husk; the resulting cake is the product of the pressing process. Animals were fed at 08h00 and 18h00, with portions controlled to allow leftovers of 15%. Water and mineral salt

were supplied *ad libitum*. After 21 d of adaptation to the nutritional management and environment, the collection and evaluation period began.

To determine the nutrient intake values, samples of the leftovers were subjected to chemical composition analysis; nutrient intake values were then calculated based on the differences remaining in the leftovers. Analysis of the samples included: dry matter (DM), organic matter (OM), mineral matter (MM), ether extract (EE), crude protein (CP), neutral detergent fiber (NDF), acid detergent fiber (ADF), cellulose (CEL), hemicellulose (HEM), lignin (LIG), protein insoluble in neutral detergent and acid detergent (PIND and PIAD), ashes insoluble in neutral and acid detergent (AIND and AIAD), and crude energy (CE), according to the methodologies of Van Soest et al. (1991), AOAC (1995), Silva; Queiroz (2002), and the recommendations of the INCT in Detmann et al. (2012). The non-fibrous carbohydrates (NFC) were calculated according to the method of Hall (2000):  $100 - (\text{NDF}_{\text{cp}} + \text{CP} + \text{EE} + \text{MM})$ . The total digestible nutrients were calculated as the sum of the digestible (D) fractions using the equation proposed by Weiss (1999):  $\text{TDN} = \text{CPD} + 2.25 \times \text{EED} + \text{NFC} + \text{NDFD}$ .

For four days, trained staff worked in alternate shifts to observe and assess the behavior of the test animals. For the first two days, staff evaluated ingestive behavior by quantifying the time intervals the animals spent idle, feeding, or in rumination during each 24-h period, according to the methodology described by Carvalho et al. (2007). Analysis with the average test showed no statistical difference between the two days, suggesting that this was an adequate amount of time for behavioral evaluation, particularly considering that the animals had been adapted to both the controlled environment and the management routine prior to data collection. Data were computed and recorded at five-minute intervals, when all animals were evaluated for the variety of the behavior exhibited (BROOM; FRASER, 2010).

During the third and fourth days of observation, staff recorded the number of chews per bolus and the rumination time per bolus (s). When recording rumination time, a digital stopwatch was used. To obtain the averages, three observations of the ruminal bolus were made, each at a different time of day, whenever the ruminating peaks were noticed: 03h00-06h00, 11h00-13h00, and 20h00-22h00 (CIRNE et al., 2014). To obtain the daily number of ruminated boluses (NRB), the total time spent ruminating was divided by the time spent ruminating each bolus for each individual animal (POLLI et al., 1996). To determine the DM and NDF in each bolus (DM g bolus<sup>-1</sup> and NDF g bolus<sup>-1</sup>) the total consumption of each was divided by the NRB. The ingestion and rumination efficiencies, in g DM h<sup>-1</sup> and g NDF h<sup>-1</sup>, were obtained by dividing the consumption of these fractions by the time spent feeding and time spent ruminating, respectively. The methods used to obtain the values of both these and additional variables, including total chewing time (TCT) in h d<sup>-1</sup> and number of ruminating chews d<sup>-1</sup>, are described by Bürger et al. (2000).

The data were tested for normality, submitted to an analysis of variance, adjusted in regression equations, and examined by Williams' test (WILLIAMS, 1971); thus, obtaining the last (W) point of the equation, which shows no significant difference at 5% error and indicates to what extent Brazil nut cake can be included in the diet without significantly affecting the response variable.

## Results and Discussion

The co-product's chemical composition (Table 1) revealed a feed containing substantial EE levels (15%) which is explained by the mechanical method of oil extraction. It is worth mentioning that these materials may vary in their compositions between industries or even between rounds of a same industry. It is also interesting to note that of the percentage (35.03%) that is CP, an advantageous nutrient, only 2.85% is considered indigestible

(PIAD), denominated fraction C according to Licitra et al. (1996).

Table 2 shows the time animals spent in each activity and the DM and NDF intake. Addition of Brazil nut cake to the diet promoted linear reductions in the consumption of DM and NDF. However, results of the Williams' test (WILLIAMS, 1971)

showed that levels of Brazil nut cake up to 30.67% of the diet do not significantly affect g NDF ingested per day, and levels of up to 51.96% of the diet do not significantly affect the daily consumption of DM. The main factors affecting consumption were reduction in the level of NDF, fiber source, and the increasing levels of fat content in the diet.

**Table 2.** Dry matter (DM) and neutral detergent fiber (NDF) intake, time spent idling (TSI), time spent rumination (TSR) and time spent feeding (TSF) in sheep fed diets containing levels of Brazil nut cake.

Variable	Inclusion level (% DM)				CV%	Equation	P Effect	W Point	R <sup>2</sup>
	0	15	30	60					
DM g day <sup>-1</sup>	915	914	786	638	18.6	Y=944.66-5.00x	0.013	51.96	0.94
NDF g day <sup>-1</sup>	351	336	280	213	18.2	Y=358.25-2.41x	0.002	30.67	0.98
TSI minutes	614	616	639	792	10.8	Y=584+3.10x	0.002	56.19	0.86
TSR minutes	530	520	519	375	11.3	Y=554.75-2.62x	0.001	56.79	0.82
TSF minutes	296	304	282	273	13.2	Y=288.75	NS	-	-

P = Probability of effect (linear or quadratic) of the treatments; CV = coefficient of variation; R<sup>2</sup> = coefficient of determination; W = estimated point to Williams test; NS = not significant (P > 0.05).

The ruminants studied herein are capable of identifying peculiarities in their feed and then altering their consumption based on previous experience (FISHER, 2002). As a result of the increased efficiency while feeding, it was necessary to develop mechanisms to indicate whether a sufficient quantity was ingested, and to interrupt the feeding, avoiding a nutrient-supply rate above that of the animals' metabolic pathways (SILVA, 2011).

The main factor affecting the animals' feed consumption is EE level; this affects consumption either by regulatory mechanisms or due to the ruminant's limited capacity to oxidize fatty acids, and rumen fermentation may be negatively affected, especially the fermentation of fibers, when exceeding 5% of the DM (PALMQUIST; MATTOS, 2006). However, the authors added that EE levels ranging from 8% to 10% provide a good response in animals confined in high temperature regions, as it increases their energy intake. The established maximum value for lipid levels in the diet is 6% EE (JORGE et al., 2008). However, in a literature review, Hess et al. (2008) found that in some cases, additions resulting

in up to 9.4% EE in the DM might not significantly affect the nutrient's digestibility.

The NDF values in the diets during T0%, T15%, and T30% were similar, 38.21%, 37.06%, and 35.91% in DM, respectively, which explains the proximity of the average time spent ruminating (TSR), a variable that underwent a decreasing linear effect ( $P < 0.01$ ), decreasing 2.62 minutes for each 1% of Brazil nut cake (NC) included in the DM. The sharp decrease at T60% is explained by high EE levels (10.7%), which probably created conditions unfavorable for rumination, affected the rumen fermentation process, and triggered lower consumption of DM and NDF.

In addition, the reduced roughage in the studied nutritional management without supplying with balanced diet, reduces the inclusion of feed types that underlie the ruminant's ability, through its microbiota, to break down food and generate energy and microbial protein. This occurs because roughage stimulates chewing activity and reduces the animal's acid production, and is thus indispensable

in the digestive process (MERTENS, 1997). When rumination and chewing time are limited, saliva production is reduced, which can lower ruminal pH, and consequently, decrease fiber digestibility (MACEDO et al., 2007).

In this study, rumination time decreased due to lower fiber consumption and the negative influence of fat on ruminal work, becoming significant from the W point of 56.79% in DM. When feed reaches the digestive tract, the intake rate limits consumption, depending mainly on the physical, mechanical, and nutritional quality of the feed. Between meals, feed is processed by the animal at a rate dependent on the area of the digestive tract, the level of enzymatic activity, bacterial load, and feed quality, and this processing rate is often the primary factor limiting food intake (BROOM; FRASER, 2010; PEREIRA et al., 2009; VAN SOEST, 1994).

From these implications regarding digestion, it can be inferred that inclusion of NC in the diet at levels higher than 56% both induced the lowest TSR and spacing between meals, and increased the idling period, which displayed increasing linear behavior. Specifically, the time spent idling increased by 3.10 minutes  $d^{-1}$  for each 1% addition of NC, with a W point estimated at 56.19%.

The time spent feeding was not affected by addition of the Brazil nut cake ( $P > 0.05$ , average time 288.75 min). However, it is thought that although the total time devoted to feeding was similar, the number of occurrences may have been different, as animals receiving more roughage probably headed to the feedlot more often due to anatomical distension and a higher need for rumination. In this sense, the animals receiving higher quantities of the co-product in their diet would have reduced physical limitation, and may therefore have remained longer while consuming the same meal. However, as a result of other digestive barriers, they may have taken longer to return to the feedlot, reducing their total consumption of DM.

Changes in the time spent feeding and ruminating have been observed in other studies, in which variations in dietary fiber content occurred. The tendency is a linear increase in idling periods, and consequently, a reduction in time spent feeding and ruminating, as the NDF levels and/or DM consumption are reduced (BÜRGER et al., 2000; CORREIA et al., 2012; SILVA et al., 2005).

Table 3 shows the values for feed and rumination efficiency, the number of ruminated boluses, and the ruminating chews. The addition of NC in the diet had a decreasing linear effect ( $P = 0.004$ ) on the number of ruminated boluses (NRB) per day, with a reduction of at least 4.61 boluses for each cake unit included; the W point was estimated at 56.64%, at which point the reduction became significant. Despite an increase in the average NRB at T30%, the equation did not show a quadratic effect ( $P > 0.05$ ). Carvalho et al. (2008) observed a trade-off between NRB and the chewing time per bolus (CTB), in which the animals spent more time per bolus when the number of ruminated boluses was lower, probably to optimize the performance. However, in this study the change in the NRB averages was not balanced by CTB ( $P > 0.05$ ), which averaged 37.5 seconds.

The addition of NC did not affect ( $P > 0.05$ ) the number of chews per bolus, an average of 56.14 chews, although the total time (TCT), in h per  $d^{-1}$ , showed a decreasing linear effect ( $P = 0.002$ ) of 0.05 h for each 1% addition of the by-product; the W point was estimated at 56.19%. The number of chews per day displayed a quadratic behavior ( $P = 0.008$ ), with an estimated maximum value of 51.215 chews in 17.50% of NC in diet. The increased amount of fiber in the diets stimulates chewing activity (MERTENS, 1997), which justifies the higher values of TCT (h  $d^{-1}$ ) in the treatments containing higher levels of fiber. The TCT is influenced by DM consumption and dietary fiber levels, as it is determined by the sum of the time spent feeding and time spent in rumination, and those variables play the primary role in the duration of these activities.

**Table 3.** Ruminated boluses, ruminating chews, feed and rumination efficiency in sheep fed diets containing levels of Brazil nut cake.

	Inclusion level (% DM)				CV%	Equation	W Point	P Effect	R <sup>2</sup>
	0%	15%	30%	60%					
Ruminal boluses									
n° dia <sup>-1</sup>	858.3	830.4	888.0	576.9	14.5	Y=909.34-4.61x	56.64	0.004	0.68
seconds	37.6	38.0	35.8	38.6	13.2	Y=37.5	-	NS	-
Rumination chews									
h dia <sup>-1</sup>	13.8	13.7	13.4	10.8	9.3	Y=14.27-0.05x	56.19	0.002	0.86
n° bolo <sup>-1</sup>	57.0	57.9	59.7	49.9	12.9	Y=56.14	-	NS	-
n° dia <sup>-1</sup>	48.971	47.334	52.194	29.088	13.4	Y=47545+419.4x-12x <sup>2</sup>	-	0.008	0.92
Feed efficiency (g h <sup>-1</sup> )									
MS	192.7	181.3	172.2	143.7	25.6	Y=172.5	-	NS	-
FDN	73.8	66.5	61.3	48.0	24.9	Y=73.54-0.42x	56.33	0.045	0.99
Rumination efficiency (g h <sup>-1</sup> )									
MS	104.2	106.2	91.4	106.0	11.7	Y=101.95	-	NS	-
FDN	39.9	39.0	32.6	35.5	11.9	Y=36.76	-	NS	-

P = Probability of effect (linear or quadratic) of the treatments; CV = coefficient of variation; R<sup>2</sup> = coefficient of determination; W = estimated point to Williams test; NS = not significant (P > 0.05).

The feeding efficiency (g DM h<sup>-1</sup>) and rumination efficiency, expressed in both g of DM and g of NDF, were not affected (P > 0.05) by the addition of the Brazil nut cake, which is explained by the fact that in the treatments that showed lower consumptions, the animals spent less time ruminating, whereas in the treatments that showed higher consumption, the animals compensated for lower efficiency by ruminating for longer periods.

The feeding efficiency, expressed as g NDF h<sup>-1</sup>, showed a decreasing linear effect (P = 0.04) of 0.42 g h<sup>-1</sup>, with the W point at 56.33%. The response was reduced due to reduced NDF consumption by the animals. According to Carvalho et al. (2008), feeding and ingestion efficiencies are affected primarily by the animals' consumption, which in turn creates implications for the time spent during ingestive activities, rumination, and idling.

## Conclusions

The Brazil nut cake, at levels greater than 30.67% of the corn-silage based diet's DM, has a negative effect, particularly on NDF consumption.

With regard to ingestive behavior, inclusion of the Brazil nut cake at levels greater than 56% particularly affects the time spent ruminating, the number of ruminated boluses in a day, and the total chewing time. This is primarily due to the levels of ether extract and the diet's source of fiber.

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