

Substitution of corn meal with dry brewer's yeast in the diet of sheep[□]

Sustitución de harina de maíz por residuos secos de cervecería en la dieta de ovejas

Substituição do farelo de milho por resíduo seco de cervejaria na dieta de ovinos

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Summary

Background: dry brewery residue can be an important tool for reducing the cost of concentrated feed for sheep. **Objective:** to evaluate the intake, digestibility and feeding behavior of sheep fed dry brewery residue. **Methods:** twenty lambs of undefined breed, with 30.33 ± 4.94 Kg body weight and seven months average age were used. Animals were fed diets containing 50% forage (Tifton-85 hay) and 50% concentrate. Treatments consisted of 0, 33, 66, and 100% corn meal substitution with dry brewery residue. Data were subjected to analysis of variance and regression analyses using a 5% significance level. **Results:** dry matter intake based on average body weight was 3.40% and showed no significant variation with the substitution level. Diet composition did not affect ($p>0.05$) intake of crude protein, ether extract, ash, lignin and total digestible nutrients (71.26%). Substitution of corn meal with dry brewery residue did not affect nutrient digestibility ($p>0.05$). Feeding behavior, time spent feeding and ruminating were not affected ($p>0.05$) by the substitution; however, idling time was quadratically related to the percentage of dry brewery residue that was substituted for corn meal, with an inflection point at 41.58% replacement. **Conclusion:** dry brewery residue can replace up to 100% corn meal without negative effects on intake, digestibility or the feeding behavior of sheep.

Keywords: *alternative foods, behavior, byproducts, digestibility, intake.*

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Resumen

Antecedentes: el uso del residuo seco de cervecería en la dieta de ovejas puede ser una herramienta importante para reducir el costo del alimento concentrado. **Objetivo:** evaluar el consumo, digestibilidad y el comportamiento de alimentación de los ovinos alimentados con residuos secos de cervecería. **Métodos:** se utilizaron 20 corderos sin raza definida, con un peso corporal de $30,33 \pm 4,94$ Kg y una edad promedio de siete meses, alimentados con dietas que contenían 50% de forraje consistente de 85% de Tifton de heno y 50% de concentrado. Los tratamientos consistieron en la sustitución de maíz por residuos secos de cervecería en las proporciones de 0, 33, 66 y 100%. Los datos fueron sometidos a análisis de varianza y regresión de prueba suponiendo un nivel de significancia del 5%. **Resultados:** el consumo de materia seca con respecto al peso corporal promedio fue de 3,40% sin mostrar alguna variación significativa. Para las ingestas de nutrientes como proteína cruda, extracto de éter, materia mineral y lignina no hubo diferencia significativa con respecto a la dieta ($p>0,05$), así como para el consumo de nutrientes digeribles totales (71,26%). Con respecto al coeficiente de digestibilidad de los nutrientes en la dieta, se encontró que ninguna de las variables sufrió efecto significativo con la sustitución de la harina de maíz por residuos secos de cervecería ($p>0,05$). El comportamiento de alimentación, el tiempo invertido en la alimentación y la rumia, no se vieron afectados ($p>0,05$) por la sustitución en la dieta, sin embargo, el tiempo de ocio tuvo un comportamiento cuadrático con punto de inflexión a la sustitución en 41,58%. **Conclusión:** el residuo seco de cervecería puede reemplazar hasta en un 100% la harina de maíz, sin causar efectos negativos sobre el consumo, la digestibilidad o el comportamiento alimenticio de las ovejas.

Palabras clave: alimentos alternativos, comportamiento, digestibilidad, ingestión, subproductos.

Resumo

Antecedentes: a utilização do resíduo seco de cervejaria na dieta de ovinos pode ser uma importante ferramenta para reduzir os custos com alimentação concentrada. **Objetivo:** avaliar o consumo e a digestibilidade do resíduo seco de cervejaria bem como o comportamento ingestivo de ovinos submetidos a esta dieta. **Método:** foram utilizados 20 cordeiros sem padrão racial definido, com peso corporal de $30,33 \pm 4,94$ Kg e idade média de sete meses, submetidos a dietas contendo 50% de volumoso constituído por feno de Tifton 85 e 50% de concentrado. Os tratamentos foram constituídos dos níveis de substituição do milho por resíduo seco de cervejaria nas proporções de 0, 33, 66 e 100%. Os dados obtidos foram submetidos à análise de variância e teste de regressão assumindo 5% de significância. **Resultados:** o consumo de matéria seca em função do peso corporal médio foi de 3,40%, não sofrendo variação significativa. Para os consumos de nutrientes como proteína bruta, extrato etéreo, matéria mineral e lignina, os resultados também não foram significativos ($p>0,05$) assim como para o consumo de nutrientes digestíveis totais (71,26%). Com relação aos coeficientes de digestibilidade dos nutrientes das dietas, verificou-se que nenhuma das variáveis estudadas sofreu efeito significativo com a substituição do milho pelo resíduo seco de cervejaria ($p>0,05$). O comportamento ingestivo e os tempos despendidos em ingestão e ruminação, não foram influenciados ($p>0,05$) pelos níveis de substituição nas dietas, entretanto o tempo de ócio teve um comportamento quadrático com ponto de inflexão em 41,58% de substituição. **Conclusão:** o resíduo seco de cervejaria pode substituir o farelo de milho em até 100% sem promover efeitos negativos no consumo, digestibilidade e comportamento ingestivo de ovinos.

Palavras chave: alimentos alternativos, comportamento, coprodutos, digestibilidade, ingestão.

Introduction

Small ruminants are able to utilize lingo-cellulosic materials and convert them to products of high nutritional value (meat, milk, wool, hide, and manure). For an animal to achieve the desired performance, a rich diet and practices that optimize welfare and performance are essential (Aguayo-Ulloa *et al.*, 2014). These diets are needed for lambs fattened in arid environments where they experience chronic stress. Even in ruminants with the ability to use low quality,

food an unbalanced diet directly influences feeding behavior, performance, and production (Martins *et al.*, 2012). Therefore, it is critically important to assess feed alternatives because feed limitation restricts production due to frequent shortages of vegetation during the dry season and the high cost of feeds.

Agro-industrial byproducts have an important economic role as supplements, and are often responsible for the viability of the system; however, the use of such byproducts in animal diets must

be carefully considered because their nutritional characteristics and availability may limit their use (Borja *et al.*, 2010; Ribeiro *et al.*, 2011; Moreira *et al.*, 2014). In particular, the fiber content of such byproducts (Torreão *et al.*, 2014) may limit intake, digestibility and animal performance (Sun and Oba, 2014).

Dry brewery residue is widely available. It was considered to be an environmental problem until recently, given that approximately 25% of the material used in the brewing process becomes residual byproducts (Gilaverte *et al.*, 2011).

Feed value is based not only on the sum of its components or nutrients but also on the overall quality of food and the extent to which it can be digested, assimilated and utilized by animals. Although few data are available concerning sheep feedlots, feed value is currently based on the difference in energy content between waste byproducts and corn meal, which is the source of energy by default (Castro-Perez *et al.*, 2014). Thus, the use of dry brewery residue in sheep diets has been hypothesized to increase intake and digestibility without negatively affecting feeding behavior. The use of this material may become important for reducing production costs. Thus, this study aimed to evaluate the optimal substitution level of corn meal with dry brewery residue in the diet of sheep.

Materials and methods

Ethical considerations

This study was conducted in strict accordance with the recommendations presented in the Guide for the National Council for Animal Experiments Control (CONCEA). The protocol was approved by the Ethics Committee on Animal Experimentation of the Federal University of Bahia (Bahia State, Brazil) (Permit Number: 17-2014).

Animals

This study was conducted at the Experimental School of Veterinary and Animal Science of the Federal University of Bahia, located in Salvador, Bahia (Brazil). Twenty non-castrated, dewormed sheep of undefined breed (SPRD) with 30.33 ± 4.94

Kg initial body weight and aged between 6 and 8 months were used. Animals were individually housed in 1.0 m² pens with suspended wooden floors and fitted with feeders and drinkers.

Experimental feeds and chemical analyses

Dietary treatments consisted of 0, 33, 66, and 100% replacement of corn meal with dry brewery residue. Concentrates were formulated with corn meal, soybean meal and dry brewery residue. The forage:concentrate ratio was 50:50, and we used Tifton-85 hay (*Cynodon* sp.) as forage (Table 1).

Diets were offered twice daily in the form of a total mixed ration (TMR) at 9 and 16 h and adjusted to allow 10% leftovers. Individual intake per animal is expressed in g/animal/day, % of body weight and g/ Kg^{0.75} (Brody, 1945).

The experiment lasted 25 days, including a 10-day pre-trial period for adaptation to the diets, facilities and management, and 15 days of data collection. Total fecal excretion was collected from day 20 to 25 to estimate apparent digestibility. Collection from individual animals during each period was performed using bags (Apack, Salvador City, Bahia State, Brazil) that were fixed to each animal. Feces were collected at the end of each day and stored in a freezer at -10 °C until further analysis.

Weighed samples of feed and leftovers from each animal were collected during the experimental period (from 10th to 25th days). Samples were packed in plastic sacks, labeled, and stored in a freezer at -10 °C until analyzed in the laboratory. Average water intake was daily measured throughout the experimental period using five-liter graduated drinkers (bucket type) to calculate volume consumed.

Samples of feed scraps and feces were pre-dried at 55 °C for 72 hours, ground using a Wiley mill (Tecnal, Piracicaba City, São Paulo State, Brazil) with a 1 mm sieve, stored in air-tight plastic container (ASS, Ribeirão Preto City, São Paulo State, Brazil), and sealed properly until laboratory analysis for concentration of DM (Method 967.03 - AOAC, 1990), ash (Method 942.05 -AOAC, 1990), CP (Method 981.10 - AOAC, 1990), and

Table 1. Chemical composition of ingredients (g/Kg DM) used in sheep diets.

Items	Ingredients			
	Corn meal	Soybean meal	Dry brewery residue	Tifton-85 hay (<i>Cynodon</i> sp.)
Dry matter	880.4	900.4	850.6	950.6
Crude protein	90.7	420.5	120.2	70.4
Neutral detergent fiber	110.3	100.9	510.2	800.8
Acid detergent fiber	40.6	100.7	410.9	450.0
Ether extract	30.0	20.6	30.8	6.2
Ash	10.4	50.7	70.1	60.2
Acid detergent lignin	10.0	8.5	60.7	90.2
Non-fiber carbohydrates	700.8	360.4	560.1	20.2
Organic matter	860.9	840.8	920.3	890.4

EE (Method 920.29 - AOAC, 1990). The NDF content was quantified using Van Soest *et al.* (1991) methodology with modifications proposed in the Ankom device manual (Ankom Technology Corporation, Macedon, New York, US). The ADF content was analyzed according to the method of Goring and Van Soest (1970). Acid detergent lignin (ADL) was quantified according to method 973.18 (AOAC, 2002), in which the ADF residue was treated with 72% sulfuric acid. The proportion of ingredients and chemical composition are shown in Table 2.

Total carbohydrate (TC) values were determined using the equation described by Sniffen *et al.* (1992): $TC = 100 - (\%CP + \%EE + \%Ash)$. In addition, the levels of non-fiber carbohydrates (NFC) were determined using the following equation: $NFC = TC - NDF$ (NRC, 2001). To calculate intake of total digestible nutrients (TDN) we used the formula proposed by Sniffen *et al.* (1992): $TDN = (iCP - fCP) + 2.25 (iEE - fEE) + (iCHO - fCHO)$, in which iCP , iEE , and $iCHO$ represent the intake of CP, EE, CHO, respectively, while fCP , fEE , and $fCHO$ represent the excretion of CP, EE, and CHO, respectively.

Ingestive behavior

Behavioral activity was assessed by observing each animal. Trained observers were positioned to ensure that they interfered as little as possible with

animal behavior. Observers recorded the time animals spent on activities of eating, ruminating and idling in five-minute intervals over a 24-hour period, as described by Johnson and Combs (1991). Artificial lighting was used during the night to facilitate feeding behavior observations.

Statistical analyses

We used a randomized complete block design for performance and behavior tests, with four treatments and five blocks defined according to the initial weight of the animals, using the following model:

$$Y_{ijk} = M + B_i + D_j + E_{ij}$$

Where:

M = average.

B_i = effect of the block ($i = 1$ to 5).

D_j = effect of supplementation with dry residue brewer (0, 33, 66, and 100%; $j = 1$ to 4).

E_{ij} = error associated with the ij observation.

Block was included as a random effect. The variables were analyzed using the MIXED procedure of SAS[®] (1999) statistical software, version 8.2 (SAS Inst. Inc, Cary, NC, USA). The averages were obtained using the LSMEANS command. The effects

Table 2. Proportions and chemical composition (g/Kg DM) of ingredients in the experimental diets.

Ingredients	Level of substitution by dry brewery residue			
	0%	33%	66%	100%
Corn meal	240.0	160.0	80.0	0.0
Soybean meal	240.5	240.5	240.5	240.5
Dry brewery residue	0.0	80.0	160.0	240.0
Mineral mix ¹	10.5	10.5	10.5	10.5
Tifton-85 hay (<i>Cynodon sp.</i>)	500.0	500.0	500.0	500.0
Nutrient				
Organic matter	932.1	927.8	923.6	923.2
Dry matter	910.8	910.8	910.8	910.7
Crude protein	164.6	169.2	168.6	170.6
Neutral detergent fiber	458.2	490.1	522.0	553.9
Acid detergent fiber	262.9	292.7	322.5	352.3
Ether extract	16.7	17.4	18.1	18.7
Ash	48.6	53.2	57.8	62.3
Acid detergent lignin	50.7	55.2	59.7	64.2
Non-fiber carbohydrates	670.7	666.2	658.7	659.8
Total carbohydrates	270.1	219.0	246.6	234.8
Digestible energy	20.4	20.4	20.5	20.5

¹The mineral mixture per Kg was as follows: 190 g calcium, 73 g phosphorus, 44 g magnesium, 62 g sodium, 92 g chloride, 30 g sulfur, 1,350 mg zinc, 340 mg copper, 940 mg manganese, 1,064 mg iron, 3 mg cobalt, 16 mg iodine, 18 mg selenium and 730 mg fluoride.

of level of dry brewer residue substitution in the diets were evaluated using linear and quadratic orthogonal polynomials. The observed effects were declared significant when $p < 0.05$.

Results

Total and Kg/day dry matter intake, as well as percentage of body weight (g/Kg) of metabolic weight did not change ($p > 0.05$) with the level of dry brewery residue substitution (Table 3).

Although no differences were observed among treatments, dry matter intake in body weight (% and g/Kg^{0.75}) showed increases of 17 and 16%, respectively, for the group that received no residue relative to the group fed 66% dry brewery residue. The maximum DM intake was 1.10 Kg/day and was observed with 66% dry brewery residue inclusion,

which corresponded to 3.65% body weight. Water intake was not affected by the quantity of dry brewery residue ($p > 0.05$). However, there was a 25% decrease between the treatment with the highest level (100%) of brewery residue and the control treatment (no dry brewery residue). The average water intake was 2.5 liters per day. As shown in Table 2, the composition of the diet was not altered in terms of dry matter and digestible energy. Average crude protein intake was 0.27 Kg/day, neutral detergent fiber intake was 0.53 Kg/day, ether extract was 0.01 Kg/day, non-fiber carbohydrate intake was 0.15 Kg/day, total carbohydrate intake was 0.68 Kg/day, and organic matter was 0.95 Kg/day for animals fed dry brewery residue.

Digestibility coefficients (Table 4) showed no significant differences for substitution of corn meal with dry brewery residue ($p > 0.05$).

Table 3. Nutrient intake in sheep fed diets containing dry brewery residue as a replacement for corn meal.

Nutrients intake	Level of substitution				CV (%)	Regression equations	P-value
	0%	33%	66%	100%			
(Kg /day)							
Dry matter	0.98	1.04	1.10	1.05	22.35	$\hat{Y} = 1.04$	0.81
Crude protein	0.27	0.28	0.28	0.28	13.23	$\hat{Y} = 0.28$	0.83
Neutral detergent fiber	0.50	0.55	0.57	0.53	18.57	$\hat{Y} = 0.54$	0.75
Ether extract	0.01	0.01	0.01	0.01	26.84	$\hat{Y} = 0.01$	0.68
Non-fiber carbohydrates	0.14	0.14	0.16	0.17	22.54	$\hat{Y} = 0.15$	0.87
Total carbohydrates	0.64	0.68	0.72	0.69	21.16	$\hat{Y} = 0.68$	0.76
Organic matter	0.89	0.96	1.01	0.97	16.91	$\hat{Y} = 0.96$	0.8
(% of body weight)							
Dry matter	3.04	3.44	3.65	3.47	22.35	$\hat{Y} = 3.40$	0.81
(g/Kg^{0.75})							
Dry matter	71.89	80.42	85.43	81.32	11.38	$\hat{Y}=79.76$	0.75
(% in total dry matter)							
Total digestible nutrients	70.88	71.54	71.51	71.12	22.46	$\hat{Y} = 71.26$	0.84
(L/day)							
Water (H ₂ O)	2.80	2.60	2.50	2.10	25.03	$\hat{Y} = 2.51$	0.87

Data are presented as means; coefficients of variation (CV); adjusted regression equations (RE) and significance (P).

Table 4. Nutrient digestibility in sheep fed diets containing dry brewery residue as a replacement for corn meal.

Digestibility	Substitution level				CV (%)	Regression equations	P-value
	0%	33%	66%	100%			
Dry matter	69.30	69.78	69.53	69.11	2.72	$\hat{Y} = 69.43$	0.67
Crude protein	77.15	76.69	76.91	79.95	3.18	$\hat{Y} = 77.68$	0.16
Neutral detergent fiber	68.26	68.90	67.39	64.35	6.62	$\hat{Y} = 67.22$	0.22
Acid detergent fiber	59.01	61.90	63.53	59.79	21.32	$\hat{Y} = 61.06$	0.20
Ether extract	50.40	53.88	52.26	52.88	8.28	$\hat{Y} = 52.36$	0.78
Non-fiber carbohydrates	70.74	70.58	70.06	68.50	2.76	$\hat{Y} = 69.97$	0.37
Total carbohydrates	78.22	78.46	79.81	82.19	8.68	$\hat{Y} = 79.67$	0.77
Organic matter	71.60	71.54	71.51	71.12	2.25	$\hat{Y} = 71.44$	0.91

Data are presented as means; coefficients of variation (CV); and adjusted regression equations (RE); data were significant when $p < 0.05$.

The inclusion of brewery residue in the diets did not affect duration of feeding and idling times (Table 5). However, the ruminating time (min/day) was a quadratic function of the level of substitution

($p < 0.05$). There was a reduction in ruminating time at the 33% substitution level and an increase in ruminating time at the 66 and 100% levels.

Table 5. Intake behavior of sheep fed diets containing dry brewery residue as a replacement for corn meal.

Variables	Level of substitution				CV (%)	P-value	
	0%	33%	66%	100%		Linear	Quadratic
Feeding (min/day)	261	279	279	258	12.08	0.8917	0.1989
Ruminating (min/day)	630	590	609	662	10.77	0.2141	0.0326*
Idling (min/day)	554	576	557	525	7.14	0.3868	0.3259
<i>Regression equation for variables</i>							
Ruminating (min/day)	$\hat{Y} = 0.02077 x^2 - 1.72985 x + 628.61142$						

Data are presented as means; coefficients of variation (CV); and adjusted regression equations (RE); data were significant when $p < 0.05$.

Discussion

The similar intake observed for all the treatments stemmed from the similarity in composition of the experimental diets. Thus, an increase of 12% lignin composition in the diet resulted in an increase in the contents of NDF and ADF and a consequent increase in dry matter intake in the form of body weight percentage as $\text{g/Kg}^{0.75}$, although this difference was not detected by the statistical test due to variability in the data. Dry matter intake may be influenced by high concentration of dietary fiber, which may limit the ingestive ability of the animals due to rumen filling. However, in this study, fiber intake had no effect on DM intake, which demonstrates that physical characteristics of fiber had no effect on ingestive ability, and the proportion of dietary fiber did not affect physiological functioning of the rumen.

Nutrient intake (CP, NDF, ADF, and EE) did not differ among diets. This can be explained by the formation of dry brewery residue from barley, which has a high content of non-structural carbohydrates. Therefore, corn meal could be replaced without altering the nutritional content of the diets. With respect to water intake, there was a trend of decreased intake after replacing corn meal with dry brewery residue. However, this decrease was not significant, thus confirming that animals accepted the substitution of corn meal with dry brewery residue and that dry brewery residue did not alter any of the physiological standards that affect water intake. The inclusion of dry brewery residue in the concentrated feed is a viable alternative to supply energy for sheep. In addition to providing a source of yeast (*Saccharomyces cerevisiae* and *S. uvarum*) with saccharification enzymes (Wang

et al., 2014), the residue also provides a flavor that improves palatability, despite having no influence on intake based on the findings. Data show that the dry brewery residue is accepted by sheep at similar rates to corn meal. All treatments resulted in total ingested water that exceeded 0.80 Kg/day (NRC, 2007), recommended for sheep by international committees.

Digestibility coefficients did not change with the inclusion of dry brewery residue. In an experiment with cattle, Geron et al. (2008) observed results similar to those found here: the inclusion of up to 24% brewery residue in the diet did not alter nutrient digestion, ruminal fermentation, nor the efficiency of microbial synthesis. The CP exhibited default behavior with all treatments. Protein composition of dry brewery residue had high values for the B2 and B3 fractions (Pereira et al., 1998) and low ruminal degradability, which together increases amino acid intake in ruminants, especially the intake of essential amino acids.

With respect to NDF and ADF digestibility, the quantities included in the diets demonstrate that the large amount of NFC in brewery residue may facilitate a greater intake of physically effective fiber. More relevant to overall digestibility is the digestibility of fiber itself and its overall proportion in the feed. Even if fiber digestibility is low, the fact that feed has less NDF makes it more digestible. Thus, an increase in OM lignin digestibility is correlated with elevated levels of NFC because the cellular content is close to 100% true digestibility (Mizubuti et al., 2014).

Substitution of corn meal with dry brewery residue did not significantly alter intake and digestibility of NDF, ADF, NFC, and TC, demonstrating that

animals regulate the digestion rate of proteins and carbohydrates, keeping TDN intake stable at an average of 71.26%. According to Cabral *et al.* (2003), the highest level of CNF, represented by fractions A + B1, theoretically increases TDN content because these carbohydrates are almost totally available to ruminants. However, in the present study, NFC levels reflected TDN level. This inconsistency may have occurred because these higher levels were not sufficient to raise the energy content based on TND calculation, which considers other nutrients capable of generating energy, such as carbohydrates and digestible proteins (Brochier and Carvalho, 2008).

Time spent ruminating depended on the proportion of dry brewery residue in the diet, as indicated by a significant quadratic effect. Although DM and NDF values -important variables to predict rumination- remained constant for all treatments, more time was spent on rumination when 41.58% of corn meal was replaced by dry brewery residue, as illustrated by the value of 592.64 min/day. The results are consistent with standard ruminant feedlot behavior.

In conclusion, dry brewery residue can completely replace corn meal in the total mixed ration or in the concentrate without negative effects on intake, digestibility or feeding time in sheep.

Conflicts of interest

The authors declare that they have no conflicts of interest with regard to the work presented in this paper.

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