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Partial replacement of corn by soybean hulls in high-grain diets for feedlot sheep

Abstract – The objective of this work was to evaluate the partial replacement of corn grains by soybean hulls, in high-grain diets, and its effect on the feeding behavior and ruminal fermentation parameters of feedlot sheep. Eight rumen-cannulated crossbred sheep were assigned to four treatment groups composed of a control, to fed a diet of 50% corn silage and 50% concentrate, and to three groups fed high-grain diets, as follows: 85% corn (Diet85), 75% corn plus 10% soybean hulls (Diet75), and 65% corn plus 20% soybean hulls (Diet65). High-grain diets reduced rumination time and provided a higher degradation of feed dry matter, a higher-propionic acid concentration, a lower-acetic acid concentration, a lower methane and carbon dioxide gas production, and a lower ruminal pH value. Diet65, with 20% soybean hulls, promoted an increase in the acetic acid proportion, with a reduction in the CO_2:CH_4 ratio. At the end of in vitro incubation, the pH value was higher for the control diet, and it was lower for Diet85. The inclusion of soybean hulls does not change methane production and dry matter degradation, and it can be used to substitute up to 20% of corn grain in high-grain diets.

Index terms: carbon dioxide, dry matter degradation, ingestive behavior, methane gas, short-chain fatty acids.

Substituição parcial de milho por cascas de soja em dietas de alta percentagem de grãos para ovinos confinados

Resumo – O objetivo deste trabalho foi avaliar a substituição parcial de milho por cascas de soja, em dietas de alta percentagem de grãos, e seu efeito sobre o comportamento alimentar e os parâmetros de fermentação ruminal em ovinos confinados. Oito carneiros com rúmens canulados foram distribuídos em quatro grupos experimentais, compostos de um controle, alimentado com uma dieta de 50% de silagem de milho e 50% de concentrado, e em três grupos alimentados com as seguintes dietas com alta percentagem de grãos: 85% de milho (Diet85), 75% de milho e 10% de cascas de soja (Diet75), e 65% de milho e 20% de cascas de soja (Diet65). Os tratamentos de alta percentagem de grãos reduziram o tempo de ruminação e proporcionaram maior degradação da matéria seca do alimento, maior concentração de ácido propiônico, menor concentração de ácido acético, menor produção de gás metano e carbono, e menor valor de pH ruminal. A Diet65, com 20% de casca de soja, promoveu o aumento da proporção de ácido acético, com a redução da relação CO_2:CH_4 no rúmen. Ao final da incubação in vitro, o valor de pH foi maior na dieta-controle, e menor na Diet85. A inclusão de cascas de soja não altera a produção de metano nem a degradação da matéria seca e pode ser usada para substituir até 20% do milho em dietas de alta percentagem de grãos.

Termos para indexação: dióxido de carbono, degradação da matéria seca, comportamento ingestivo, gás metano, ácidos graxos de cadeia curta.

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Introduction

The use of large proportions of concentrate in ruminant diets has been a strategy for maximizing the performance of production animals (Kumar et al., 2014; Oliveira & Millen, 2014). In intensive meat production systems, such as feedlots, the use of diets with a high proportion of cereal grains is a well-known practice. However, digestive and metabolic problems, such as acidosis and bloating, are a common consequence of high-grain diets, and nutritional and management strategies should be adopted (Hernández et al., 2014).

The lack of a roughage source can reduce saliva production and impair rumination and ruminal motility (Minervino et al., 2014). The increase of grain proportion of a diet promotes ruminal fermentation with a predominance of acid pH, especially in the first hours after ingestion. This acidic pH is a consequence of the increased concentration of propionic acid in the rumen (Van Baale et al., 2004), largely due to the fermentation of cereal grains, such as corn. Drastic and prolonged pH reductions may inhibit the growth of cellulolytic and methanogenic microorganisms, which may impair feed digestibility and animal performance (Krajcarski-Hunt et al., 2002). Thus, the manipulation of ruminal fermentation through the substitution of highly fermentable ingredients is an essential nutritional strategy for animal health to maximize production performance.

Brazil has a diversity of agroindustrial by-products with great potential to be used as animal feed such as soybean hulls. Furthermore, a partial substitution of corn for soybean hulls may be a useful strategy to avoid nutritional and metabolic problems, minimizing the negative effects of excessive fermentation caused by high-grain diets, and aiding the rumen motility and buffering capacity. However, soybean hulls are high in fiber, similarly to roughage feeds, which can lead to an increase of methane production, consequently affecting the animal performance, since methanogenesis may represent a loss of up to 18% of gross dietary energy (Kozloski, 2017).

The objective of this work was to evaluate the partial replacement of corn by soybean hulls, in high-grain diets, and its effects on feed behavior and ruminal fermentation parameters of feedlot sheep.

Materials and Methods

Eight rumen-cannulated crossbred uncastrated adult Santa Inês sheep were housed in individual pens (4.5 m^2) indoors, with cement floors and individual feed bunks and waterers. The animals received four experimental diets (Table 1), as follows: a control treatment, with a diet of 50% corn silage and 50% concentrate, and three high-grain treatments. All high-grain diets contained 15% of commercial pelleted concentrate, and differed for the proportion of corn and soybean hulls: Diet85, 85% corn; Diet75, 75% corn and 10% soybean hulls; and Diet65, 65% corn and 20% soybean hulls (Table 2). Animals were given two meals a day, at 7:00 and at 19:00.

Each of the four experimental periods had a total duration of 21 days, with 15 days for diet adaptation, and 6 days for data collection. On the 16^{th} day of each experimental period, the animals' feeding behavior was evaluated every 5 min for 24 hours. The observed variables were time spent in idle, ruminating, and feeding, expressed as a percentage of 24 hours. For the determination of rumen pH, ammoniacal nitrogen (NH_3-N), and short-chain fatty acids (SCFA) concentrations, samples were collected from ruminal contents on the 20^{th} day of each experimental period at 0, 2, 4, 8, and 12 hours after feeding, in which time 0 referred to the time before morning feeding (7:00 h). The pH was measured directly in the filtered rumen fluid (in a nylon membrane, pore size 100 μm), using a digital pH meter with standard buffer solutions, at pH 4.0 and 7.0. The analysis of NH_3-N concentration was performed by distillation of the rumen fluid (13 mL of H_2O and 5 mL of 2 mol L^{-1} KOH) and titration, using 0.005 mol L^{-1} hydrochloric acid (Souza et al., 2013). For the determination of SCFA (acetic, propionic, butyric, isobutyric, valeric, and isovaleric acids), a 5 mL sample of filtered rumen fluid was diluted in 1 mL of formic acid, following the methodology described by Leventini et al. (1990) for further chromatographic reading. The concentration of SCFA was determined by injecting 0.5 μL of sample in a gas chromatograph (Trace 1300, Thermo Scientific, MA, USA), equipped with a HP-FFAP capillary column model 19091F-112 (25 m, 0.320 mm, 0.50 μm, J&W Agilent Technologies Inc., Palo Alto, CA, USA). The carrier gas was helium at a 1 mL min^{-1} flow rate. The oven temperature program was 1 min at 60°C, followed by an increase to 200°C at 5°C min^{-1} rate. The injector temperature was

270°C, and the detector temperature was 300°C. The sample was injected into a split/splitless system (split ratio 1:10).

Gas measurements were performed on the 21st day of each experimental period. The ruminal content was collected before the morning feeding and filtered to obtain the rumen fluid for in vitro incubation. Feed ingredients were grounded (1 mm), weighed (approximately 1.87 g dry matter, DM), and placed in Erlenmeyer flasks (250 mL) containing 150 mL of rumen fluid. Four Erlenmeyer flasks containing only rumen fluid were used as blanks and each experimental diet was analyzed in five replicates, totaling 24 samples (Homem Junior et al., 2015). This procedure was carried out in all experimental periods. Samples were incubated at 39°C for 24 hours, and gases produced during incubation were conducted by capillary tubes and stored in plastic containers. Therein, the gas column height (H, cm) was measured to estimate the total gas volume (TV, mL), using the equation . After determining the TV, an aliquot of 0.5 mL gas was sampled from the flasks to determine concentrations of CO_2 and CH_4, using a gas chromatograph (Trace GC Ultra, Thermo Scientific, San Jose, CA, USA). The GC was equipped with a Porapak column and a molecular sieve. The oven temperature was set at 70°C, and the injector temperature at 110°C. The carrier gas used was argon, with 25 mL min^{-1} flow.

After incubation, the pH values of samples were measured with a digital bench-top pH meter. To obtain the nondegradable incubation residues, the samples were centrifuged for 3 min at 3000 rpm, and they were subsequently dried in an oven at 105°C until a constant mass was obtained for the determination of dry matter degradation (DM Deg). Sample DM Deg was calculated using the following equation (Chaudhry & Khan, 2012): Deg = ISW – [(RW – BRW)/ISW] x 100; in which: ISW stands for the weight of the incubated sample; RW is the weight of residue; and BRW is the weight of the blank residue.

Data were analyzed as a replicated 4×4 Latin square design using Proc Mixed of SAS version 9.2. The fixed effect consisted of treatments, and random effects consisted of sheep and periods. Data of pH, NH_3-N, and SCFA were considered as repeated measures. Several covariance structures were tested, and the best one was chosen for each variable, based on the Akaike's information criterion. The degrees of freedom and tests were adjusted using the option KR. Treatment mean differences were determined by the Tukey's test, at 5% probability.

Table 1. Dry matter and crude protein content of feed ingredients (g kg^{-1} DM).

Ingredient	Dry matter	Crude protein
Corn silage(1)	330.0	80.0
Commercial concentrate(2)	880.0	180.2
Corn grain(1)	870.1	90.5
Pelleted concentrate(2)	900.0	330.3
Soybean hulls(1)	900.1	110.6

(1)Nutritional values obtained by laboratory analysis. (2)Values were informed by the company Manufaturação de Produtos para Alimentação Animal Premix Ltda.

Table 2. Ingredient and chemical composition of experimental treatments.

Item	Treatments(1)			
	Control	Diet85	Diet75	Diet65
	Ingredient composition (g kg^{-1} DM)			
Corn silage	500.0	-	-	-
Commercial concentrate	500.0	-	-	-
Corn grain	-	845.8	743.7	642.4
Pelleted concentrate	-	154.2	153.7	153.2
Soybean hulls	-	-	102.6	204.4
	Nutrient composition			
Crude protein (g kg^{-1} DM)(2)	130.1	131.6	133.7	135.8
Dry matter(1)	478.5	875.4	878.4	881.4

(1)Control diet, 50% corn silage and 50% commercial concentrate; Diet85, 85% corn; Diet75, 75% corn and 10% soybean hulls; Diet65, 65% corn and 20% soybean hulls. (2)Values were informed by the company Manufaturação de Produtos para Alimentação Animal Premix Ltda.

Results and Discussion

Animals receiving the high-grain diets, irrespectively of the inclusion of soybean hulls replacing corn grain, provided less time spent in rumination and feeding, and longer periods spent lying down idle than the animals that fed the control diet (Table 3). The reduction of more than 50% in ruminating time may be a consequence of the absence of forage source (corn silage), a fact that also affected the time spent for feeding. According to Owens & Basalan (2016), the duration of time spent ruminating and feeding is proportional to the quantity and quality of fiber floating in the ruminal raft. As high-grain diets have low-fiber and high-nonstructural carbohydrate, the consumption, fermentation, and use of these diets are faster than forage-based diets. In this way, the lower-feeding time justifies the longer-time lying down idle for animals fed high-grain diets.

Despite the feeding-time reduction, high-grain diets did not affect the dry matter intake (DMI) of the animals, except for the diet with 85% corn grains (Diet85), which showed a lower DMI than the other diets. This fact can be due to a metabolic status improvement of the animals because of the increased energy intake of starch (corn grain), with increased propionate and reduced acetate in the rumen (Table 4) that satiated the animals for energy.

The inclusion of fiber-rich ingredients in high-concentrate diets may be beneficial to improve rumination of animals according to Minervino et al. (2014), who observed that animals fed a diet of 80% forage (Coast-Cross hay) spent 46% of the day in rumination, while animals assigned to diets with 30% forage, using corn grains in the concentrate composition, spent 29% of the day in rumination, and when pelleted citrus pulps were used, the proportion of the day spent in rumination was 42%. However, in the present study, the partial replacement of corn grains by soybean hulls did not promote more rumination to the animals. This fact can explain the lower-pH values observed for high-grain diets (Table 4).

Although the feeding of high-energy diets increases propionate, and decreases acetate proportion in the rumen, as observed in this study, the lack of fiber can impair the cellulolytic activity in the rumen, and decrease salivation through the reduction of feeding and ruminating. Salivation leads to increasing ruminal pH that favors the growth of cellulolytic bacteria and protozoa (Millen et al., 2016). Growth of these microorganisms are inhibited by a pH below 6.0, which decreases the fiber digestibility in the rumen that leads to reduced feed intake (Russell & Wilson, 1996), as observed for Diet85 (Table 3). In addition, feeding a large amount of grains, mainly with high content of starch, can promote the growth of lactic acid bacteria, which reduces pH in the rumen and could lead to acidosis (Millen et al., 2016).

Table 3. Sheep behavior during 24 hours of observation(1).

Item	Treatments(2)				Probability value	Standard error
	Control	Diet85	Diet75	Diet65		
Rumination (%)	28.04a	13.94b	13.65b	13.11b	***	2.22
Idle (%)	36.08a	43.36a	42.60a	43.25a	ns	3.19
Lying down idle (%)	16.70a	29.33b	28.00b	28.42b	*	4.41
Feeding (%)	13.50a	9.58b	10.26b	10.54b	*	1.17
Drinking water (%)	5.66a	4.61a	4.81a	3.83a	ns	1.63
DMI (kg per day)(3)	1.46a	0.93b	1.23ab	1.25ab	*	0.19

(1)Means followed by equal letters do not differ by Tukey's test, at 5% probability. *, ***Significant at 5 and 1% probability; ns, nonsignificant. (2)Control diet, 50% corn silage and 50% commercial concentrate; Diet85, 85% corn; Diet75, 75% corn and 10% soybean hulls; Diet65, 65% corn and 20% soybean hulls. (3)DMI, dry matter intake.

Table 4. Ruminal fermentation parameters of feedlot sheep(1).

Item(2)	Treatments(3)				Effect		
	Control	Diet85	Diet75	Diet65	Treatment	Treatment×TI(4)	Standard error
pH	6.30a	5.69b	5.87b	5.75b	***	ns	0.06
NH_3-N (mg dL^{-1})	32.82b	38.59a	31.94b	40.52a	**	ns	1.50
SCFA (mmol L^{-1})	86.94b	97.10ab	98.56ab	110.10a	**	ns	7.67
C2 (%)	63.16a	48.11c	50.85bc	53.36b	***	ns	1.53
C3 (%)	19.90b	35.34a	37.29a	33.74a	***	ns	2.65
C4 (%)	12.67a	11.88ab	7.50c	8.57bc	*	ns	1.50
Isobut (%)	1.52a	1.17ab	1.05b	1.05b	*	ns	0.16
Isoval (%)	1.72a	2.19a	2.13a	2.11a	ns	ns	0.28
Valeric (%)	1.00b	1.29a	1.15ab	1.16ab	*	ns	0.09

(1)Means followed by equal letters do not differ by Tukey's test, at 5% probability. P, probability value. *, ***Significant at 5 and 0.1%. nsNonsignificant. (2)NH_3-N, ammoniacal nitrogen; SCFA, short-chain fatty acids; C2, acetic acid; C3, propionic acid, C4, butyric acid; isobut, isobutyric acid; isoval, isovaleric acid; valeric, valeric acid. (3)Control diet, 50% corn silage and 50% commercial concentrate; Diet85, 85% corn; Diet75, 75% corn and 10% soybean hulls; Diet65, 65% corn and 20% soybean hulls. (4)Treatment×TI, treatment × time interaction.

The lowest-NH_3-N concentrations were observed in animals fed the control diet and the Diet75 with 10% soybean hulls (Table 4). These diets have structural carbohydrates in their composition, and since the microorganisms that ferment cellulose and hemicellulose use only ammonia as a N source for microbial protein synthesis (Russell et al., 1992), it is probable that the ammonia was better utilized. Besides, for Diet65 with 20% soybean hulls, the rate of protein degradation may have exceeded the rate of carbohydrate fermentation, and large quantities of N were lost as ammonia. As to Diet85, with 85% of corn grains, there was probably no fermentative synchrony between N and energy because of a high-starch content, which may have caused no use of ammonia by microorganisms. In addition, microorganisms that ferment starch use either ammonia or amino acids as an N source (Russell et al., 1992). Nonetheless, the concentrations of ruminal NH_3-N were sufficient for bacterial growth in all treatments. In agreement with Preston (1986), the minimal concentration of 5 mg NH_3-N dL^{-1} is sufficient for microbial growth, however, the concentration should be above 10 mg dL^{-1} for increase in ruminal digestion (Leng, 1990).

Diet65 with 20% soybean hulls increased the total SCFA production in the rumen, and Diet85 e Diet75 did not differ from the control diet (Table 4). The higher inclusion of soybean hulls could explain this result, since this source of structural carbohydrate may have provided a higher concentration of acetic acid (Table 4) and, consequently, higher values of valeric acid. The high-grain diets promoted lower concentrations of butyric and isobutyric acids in the rumen, with lowest concentrations for diets with soybean hulls (Diet75 and Diet65). These lowest concentrations may be due to a greater absorption rate because when ruminal pH is low, close to 5.0, there is an increase of the absorption rate. According to Russell (1998), the increase of absorption rate with the pH reduction is greater for butyric acid, intermediate for propionic acid, and null for acetic acid. This is due to the fact that butyric acid is almost completely metabolized by the ruminal wall.

High-grain diets had higher-dry matter degradation values than the control diet, and lower methane gas (CH_4 mL g^{-1} and CH_4 mL gd^{-1}) and carbon dioxide (CO_2 mL gd^{-1}) production (Table 5). These results agree with those reported by Pedreira et al. (2013), who evaluated diets with three proportions of dietary forage, using 100, 70, and 40% of sorghum silage, and showed a reduction of CH_4 production when animals fed a diet of 100% sorghum silage. According to Doreau et al. (2011), high proportion of concentrate in diets promoted a better degradation and utilization in the rumen. However, this increase is not necessarily accompanied by an increase of the production of SCFA, but always by a reduction of ruminal pH and CH_4 production, as observed in the present study (Table 4).

Animals that received the control diet with a high amount of forage had a higher amount of CH_4 production than those which received high-grain diets (Table 5). The high-grain diets reduced the CH_4 production in more than 35% (mL g^{-1}). This behavior is directly related to the proportion of acetic, propionic, and butyric acids formed. For instance, the higher proportion of propionate formed during ruminal fermentation enables H_2 capture without CO_2 formation, which does not favor the methanogenic bacteria. However, the production of acetate and butyrate liberates CO_2 and H_2, which are CH_4 precursors, into the ruminal environment (Janssen, 2010). Ingredients with high nonstructural carbohydrate may increase the proportion of propionic acid produced, thus decreasing methane production (Mitsumori & Sun, 2008), as observed in the present study (Tables 3 and 4). The lower CH_4 production can explain the lower total gas production of high-grain diets (Table 4).

The control diet promoted a lower-CO_2/CH_4 ratio than the high-grain diets; and the inclusion of soybean hulls in partial substitution to corn resulted in a decrease of the CO_2/CH_4 ratio (Table 5). This result can characterize soybean hulls as a dietary forage ingredient, with a fermentation pattern of acetic acid production (Table 4). The lower-CO_2/CH_4 ratio was due to lower CH_4 production in high-grain diets, as reported in a study by Christophersen et al. (2008), who evaluated two diets, one with high-grain percentage and other with high- roughage percentage, reporting a decrease of about 15% CH_4 production

The pH value at the end of the in vitro incubation was higher for the control diet, but it was lower for the high-grain diets without soybean hulls (Table 5). High-grain diets were effective in reducing the CH_4 and CO_2 production, possibly due to the increase of the proportion of propionic acid, and the reduction of acetic acid and rumen fluid pH. The increase of propionate

Table 5. Dry matter degradation, gas production, and pH of experimental diets after fermentation in vitro[1].

Item[2]	Treatments[3]				Probability value	Standard error
	Control	Diet85	Diet75	Diet65		
Deg (% DM)	49.76b	62.27a	58.88a	59.36a	***	1.01
Total gas (mL g^{-1})	102.88a	96.69b	95.87b	94.22b	***	1.32
CH_4 (mL g^{-1})	18.36a	13.73b	14.47b	14.41b	***	0.37
CO_2 (mL g^{-1})	69.10a	68.71a	68.52a	65.15a	ns	1.61
CH_4 (mL gd^{-1})	36.99a	22.55b	24.73b	24.53b	***	0.87
CO_2 (mL gd^{-1})	139.12a	112.82b	117.06b	110.88b	***	3.54
CO_2/CH_4	3.74d	5.00a	4.75b	4.52c	***	0.04
pH in vitro	5.52a	5.33d	5.36c	5.41b	***	0.01

[1]Means followed by equal letters do not differ by Tukey's test, at 5% probability. *, ***Significant at 5 and 0.1%. [ns]Nonsignificant. [2]Deg, ruminal dry matter degradation in vitro after 24 hours; CH_4g, methane production (mL g^{-1} DM); CO_2g, carbon dioxide production (mL g^{-1} DM); CH_4gd, methane production (mL g^{-1} DM degraded after 24 hours); CO_2gd, carbon dioxide production (mL g^{-1} DM degraded after 24 hours); pH, mean pH in vitro; CO_2/CH_4, ratio between CO_2 and CH_4. [3]Control diet, 50% corn silage and 50% commercial concentrate; Diet85, 85% corn; Diet75, 75% corn and 10% soybean hulls; Diet65, 65% corn and 20% soybean hulls.

and the decrease of ruminal pH are in accordance with the results reported by Christophersen et al. (2008). In the present study, sheep that fed diets containing 70% of oat grains had an increased production of propionate, a reduced ruminal pH, and a reduced CH_4 production in comparison to sheep fed a diet with 63% of oat straw, but a reduction of acetate production was not observed.

Conclusions

1. The use of high-grain diets reduces the methane production and rumination time.
2. Soybean hulls does not alter the feeding behavior and methane production in comparison to high-grain diets with only corn grain.
3. Soybean hulls can be used to replace up to 20% of corn grain in high-grain diets.

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References

CHAUDHRY, A.S.; KHAN, M.M.H. Impacts of different spices on in vitro rumen dry matter disappearance, fermentation and methane of wheat or ryegrass hay based substrates. **Livestock Science**, v.146, p.84-90, 2012. DOI: https://doi.org/10.1016/j.livsci.2012.01.007.

CHRISTOPHERSEN, C.T.; WRIGHT, A.-D.G.; VERCOE, P.E. In vitro methane emission and acetate:propionate ratio are decreased when artificial stimulation of the rumen wall is combined with increasing grain diets in sheep. **Journal of Animal Science**, v.86, p.384-389, 2008. DOI: https://doi.org/10.2527/jas.2007-0373.

DOREAU, M.; VAN DER WERF, H.M.G.; MICOL, D.; DUBROEUCQ, H.; AGABRIEL, J.; ROCHETTE, Y.; MARTIN, C. Enteric methane production and greenhouse gases balance of diets differing in concentrate in the fattening phase of a beef production system. **Journal of Animal Science**, v.89, p.2518-2528, 2011. DOI: https://doi.org/10.2527/jas.2010-3140.

HERNÁNDEZ, J.; BENEDITO, J.L.; ABUELO, A.; CASTILLO, C. Ruminal acidosis in feedlot: from aetiology to prevention. **The Scientific World Journal**, v.2014. art. ID 702572, 2014. DOI: https://doi.org/10.1155/2014/702572.

HOMEM JUNIOR, A.C.; EZEQUIEL, J.M.B.; PEREZ, H.L.; ALMEIDA, M.T.C.; PASCHOALOTO, J.R.; CARVALHO, V.B. de; CREMASCO, L.F.; COSTA, M.B. da. In vitro fermentation of corn silage using rumen fluid buffered or not and different sample amounts. **Ciência Rural**, v.45, p.2229-2232, 2015. DOI: https://doi.org/10.1590/0103-8478cr20140902.

JANSSEN, P.H. Influence of hydrogen on rumen methane formation and fermentation balances through microbial growth kinetics and fermentation thermodynamics. **Animal Feed Science and Technology**, v.160, p.1-22, 2010. DOI: https://doi.org/10.1016/j.anifeedsci.2010.07.002.

KOZLOSKI, V.G. **Bioquímica dos ruminantes**. 3. ed. rev. e ampl. Santa Maria: Ed. da UFSM, 2017. 212p.

KRAJCARSKI-HUNT, H.; PLAIZIER, J.C.; WALTON, J.-P.; SPRATT, R.; MCBRIDE, B.W. Effect of subacute ruminal acidosis on in situ fiber digestion in lactating dairy cows. **Journal of Dairy Science**, v.85, p.570-573, 2002. DOI: https://doi.org/10.3168/jds.S0022-0302(02)74110-6.

KUMAR, S.; CHOUDHURY, P.K.; CARRO, M.D.; GRIFFITH, G.W.; DAGAR, S.S.; PUNIYA, M.; CALABRO, S.; RAVELLA,

S.R.; DHEWA, T.; UPADHYAY, R.C.; SIROHI, S.K.; KUNDU, S.S.; WANAPAT, M.; PUNIYA, A.K. New aspects and strategies for methane mitigation from ruminants. **Applied Microbiology and Biotechnology**, v.98, p.31-44, 2014. DOI: https://doi.org/10.1007/s00253-013-5365-0.

LENG, R.A. Factors affecting the utilization of 'poor-quality' forages by ruminants particularly under tropical conditions. **Nutrition Research Reviews**, v.3, p.277-303, 1990. DOI: https://doi.org/10.1079/NRR19900016.

LEVENTINI, M.W.; HUNT, C.H.; ROFFLER, R.E.; CASEBOLT, D.G. Effect of dietary level of barley-based supplements and ruminal buffer on digestion and growth by beef cattle. **Journal of Animal Science**, v.68, p.4334-4344, 1990. DOI: https://doi.org/10.2527/1990.68124334x.

MILLEN, D.D.; PACHECO, R.D.L.; CABRAL, L. da S.; CURSINO, L.L.; WATANABE, D.H.M.; RIGUEIRO, A.L.N. Ruminal acidosis. In: MILLEN, D.D.; ARRIGONI, M.D.B.; PACHECO, R.D.L. (Ed.). **Rumenology**. Cham: Springer, 2016. p.127-156. DOI: https://doi.org/10.1007/978-3-319-30533-2_5.

MINERVINO, A.H.H.; KAMINISHIKAWAHARA, C.M.; SOARES, F.B.; ARAÚJO, C.A.S.C.; REIS, L.F.; RODRIGUES, F.A.M.L.; VECHIATO, T.A.F.; FERREIRA, R.N.F.; BARRÊTO-JÚNIOR, R.A.; MORI, C.S.; ORTOLANI, E.L. Behaviour of confined sheep fed with different concentrate sources. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v.66, p.1163-1170, 2014. DOI: https://doi.org/10.1590/1678-6366.

MITSUMORI, M; SUN, W. Control of rumen microbial fermentation for mitigating methane emissions from the rumen. **Asian-Australasian Journal of Animal Sciences**, v.21, p.144-154, 2008. DOI: https://doi.org/10.5713/ajas.2008.r01.

OLIVEIRA, C.A.; MILLEN, D.D. Survey of the nutritional recommendations and management practices adopted by feedlot cattle nutritionists in Brazil. **Animal Feed Science and Technology**, v.197, p.64-75, 2014. DOI: https://doi.org/10.1016/j.anifeedsci.2014.08.010.

OWENS, F.N.; BASALAN, M. Ruminal fermentation. In: MILLEN, D.D.; ARRIGONI, M.D.B.; PACHECO, R.D.L. (Ed.). **Rumenology**.Cham: Springer, 2016. p.63-102. DOI: https://doi.org/10.1007/978-3-319-30533-2_3.

PEDREIRA, M. dos S.; OLIVEIRA, S.G. de; PRIMAVESI, O.; LIMA, M.A. de; FRIGHETTO, R.T.S.; BERCHIELLI, T.T. Methane emissions and estimates of ruminal fermentation parameters in beef cattle fed different dietary concentrate levels. **Revista Brasileira de Zootecnia**, v.42, p.592-598, 2013. DOI: https://doi.org/10.1590/S1516-35982013000800009.

PRESTON, T.R. **Better utilization of crop residues and by-products in animal feeding**: research guidelines. 2. A practical manual for research workers. Rome: FAO, 1986. (FAO. Animal Production and Health Paper, 50/2).

RUSSELL, J.B. The importance of pH in the regulation of ruminal acetate to propionate ratio and methane production in vitro. **Journal of Dairy Science**, v.81, p.3222-3230, 1998. DOI: https://doi.org/10.3168/jds.S0022-0302(98)75886-2.

RUSSELL, J.B.; O'CONNOR, J.D.; FOX, D.G.; VAN SOEST, P.J.; SNIFFEN, C.J. A net carbohydrate and protein system for evaluating cattle diets: I. Ruminal fermentation. **Journal of Animal Science**, v.70, p.3551-3561, 1992. DOI: https://doi.org/10.2527/1992.70113551x.

RUSSELL, J.B.; WILSON, D.B. Why are ruminal cellulolytic bacteria unable to digest cellulose at low pH? **Journal of Dairy Science**, v.79, p.1503-1509, 1996. DOI: https://doi.org/10.3168/jds.S0022-0302(96)76510-4.

SOUZA, N.K.P.; DETMANN, E.; VALADARES FILHO, S.C.; COSTA, V.A.C.; PINA, D.S.; GOMES, D.I.; QUEIROZ, A.C.; MANTOVANI, H.C. Accuracy of the estimates of ammonia concentration in rumen fluid using different analytical methods. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v.65, p.1752-1758, 2013. DOI: https://doi.org/10.1590/S0102-09352013000600024.

VAN BAALE, M.J.; SARGEANT, J.M.; GNAD, D.P.; DEBEY, B.M.; LECHTENBERG, K.F.; NAGARAJA, T.G. Effect of forage or grain diets with or without monensin on ruminal persistence and fecal *Escherichia coli* O157:H7 in cattle. **Applied and Environmental Microbiology**, v.70, p.5336-5342, 2004. DOI: https://doi.org/10.1128/AEM.70.9.5336-5342.2004.