

## ***In vitro* rumen fermentation kinetics of diets containing oldman saltbush hay and forage cactus, using a cattle inoculum**

[*Cinética de fermentação ruminal in vitro de dietas contendo feno de erva-sal e palma forrageira, utilizando inóculo bovino*]

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### **ABSTRACT**

The aim of this experiment was to evaluate, by means of the semi-automated *in vitro* gas production technique, fermentation kinetics of carbohydrates and degradability of dry matter (DM) and organic matter (OM) of diets containing oldman saltbush hay levels (8.4; 18.8; 31.2 and 48.3%) associated to forage cactus *in natura*. Pressure readings of the gases were done with a pressure transducer at 2, 4, 6, 8, 10, 12, 17, 20, 24, 28, 34, 48, 72 and 96h post-inoculation. The rumen kinetics was described by the following parameters: maximum potential of gas production, lag time and production rates of gas (k), fibrous carbohydrates (FC) and non-fibrous carbohydrates (NFC). It could be observed that the addition of oldman saltbush hay to the diets promoted a quadratic effect in the production of gases originated from NFC. However, there was no significant effect on the production of gases originated from FC and on production rates of gases from NFC and FC. The degradability of DM and OM did not differ due to the addition of oldman saltbush hay. The use of 8.4% hay and 74.9% forage cactus promoted the maximum potential of production of gases from the fibrous fraction of diets containing cactus and oldman saltbush hay.

Keywords: *Atriplex nummularia*, degradability, forages, *Opuntia ficus indica*

### **RESUMO**

O objetivo deste experimento foi avaliar, por meio da técnica *in vitro* semiautomática de produção de gases, a cinética de fermentação dos carboidratos e a degradabilidade da matéria seca (MS) e da orgânica (MO) de dietas contendo níveis de feno de erva-sal (8,4; 18,8; 31,2 e 48,3%) associado à palma forrageira *in natura*. As leituras de pressão dos gases foram feitas com transdutor de pressão às 2, 4, 6, 8, 10, 12, 14, 17, 20, 24, 28, 34, 48, 72, 96 horas pós-inoculação. A cinética ruminal foi descrita pelos parâmetros: potencial máximo de produção de gases, “lag time” e taxa de produção de gases (k) dos carboidratos fibrosos (CF) e não fibrosos (CNF). Verificou-se que a adição de feno de erva-sal às dietas promoveu efeito quadrático na produção de gases provenientes dos CNF, em que o menor valor observado (136,48mL) foi encontrado com a adição de 38,25% de feno, e o maior valor, com a adição de 8,4% de feno e 74,9% de palma forrageira. O tempo de latência teve comportamento quadrático em função da adição de feno. No entanto não houve efeito significativo na produção de gases provenientes dos CF (média de 111,6mL) e nas taxas de produção de gases dos CNF e CF que apresentaram média de 0,090h<sup>-1</sup> e 0,028h<sup>-1</sup>, respectivamente. A degradabilidade da MS e MO não diferiram em função da adição de feno de erva-sal e apresentaram médias de 85,8 e 90,9%, respectivamente. O uso de 8,4% de feno e 74,9 de palma forrageira propiciou o máximo potencial de produção de gases da fração fibrosa de dietas contendo palma e feno de erva-sal.

Palavras-chave: *Atriplex nummularia*, degradabilidade, forragens, *Opuntia ficus indica*

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## INTRODUCTION

Halophyte plants are forage sources that are commonly utilized in many parts of the world. Among these plants, oldman saltbush (*Atriplex nummularia*) has been used to sustain livestock activities in regions where animal feed is scarce.

Oldman saltbush is tolerant to salinity and has the capacity to accumulate significant levels of salts within its tissues, allowing it to adapt to arid and semi-arid regions where the average annual rainfall ranges from 200 to 400mm (Le Houérou, 1993). In these regions, the production of dry matter from fresh material is approximately 3.4ton/ha/year during low-rainfall conditions (390 mm/year) and 8.0ton/ha/year when irrigated (El Shaer, 2010).

Several studies have previously evaluated the nutritional quality of oldman saltbush and its usage in the feeding of small ruminants, particularly sheep. El-Waziry (2007) reported that despite the satisfactory crude protein content detected within its leaves, 126-144.5g.kg<sup>-1</sup> in dry matter (DM), *Atriplex nummularia* is only moderately digestible, exhibiting low digestibility of ether extracts and containing high levels of oxalate and minerals and low levels of soluble carbohydrates. In addition, the fresh leaves of this forage source contain high levels of lignin (85g.kg<sup>-1</sup>) in the DM (El-Waziry, 2007). Thus, the identification of a forage source that could be combined with oldman saltbush to diminish its negative characteristics is urgently needed.

The forage cactus (*Opuntia ficus indica*) is a member of the cactaceae that is adapted to semi-arid regions and that contains low levels of dry matter, crude protein and fiber, and high levels of soluble carbohydrates. The advantages of utilizing cactus as a forage source in conjunction with oldman saltbush have been described by Ben Salem *et al.* (2002), who reported the possibility of minimizing the negative effects of using oldman saltbush as an exclusive feed by combining it with forage cactus. However, few specific studies have evaluated the positive effects that such a combination confers in terms of rumen fermentation kinetics.

The semi-automated *in vitro* gas production technique has been successfully utilized in this type of study. Apart from the characterization of rumen kinetics, the gas production technique enables the study of anti-nutritional factors

present in the feedstuffs. In contrast with *in situ* or *in vitro* techniques based on gravimetric determinations, the gas production technique does not allow for the solubilization of the anti-nutritional factors; thus, there is no risk of their being quantified as degraded dry matter. Even though the volume of gas produced when feeds are incubated *in vitro* with buffered rumen fluid is only a reflection of rumen events, it has been related to whole tract digestibility and has also been used as a means of estimating the metabolisable and net energy content of feeds (Rymer and Givens, 2002); then, the *in vitro* gas production could be used as a tool to evaluate nutritive value and compare different forage sources as a preliminary study before the realization of field animal trial.

In this context, this study aimed to evaluate the range and kinetics of the rumen fermentation of diets containing oldman saltbush hay combined with forage cactus by means of the semi-automated *in vitro* gas production technique.

## MATERIALS AND METHODS

The study was conducted at Semiarid Embrapa, located in Petrolina-PE, Brazil.

The treatments evaluated corresponded to diets containing 8.4, 18.8, 31.2, and 48.3% oldman saltbush (*Atriplex nummularia*) hay (% in DM) in combination with forage cactus (*Opuntia ficus-indica* Mill) (Table 1). Concentrates containing different levels of urea: ammonium sulfate (9:1), ground corn and soybean meal were included to ensure that all diets contained equal amounts of protein and energy.

Oldman saltbush was obtained from growing areas in Semiarid Embrapa. The material collected was weighed, dried in the sun and then chopped. Forage cactus were obtained from the experimental field of Semiarid Embrapa and then processed in a stationary chopper that was appropriate for forage cactus. Before the formulation of the complete diets, the concentrate ingredients were pre-mixed (Table 1) and then added to the oldman saltbush and cactus in pre-established amounts. Samples of the diet mixes and feeds were collected, dried in a ventilation oven at 55 °C for 72 hours, ground in a Willey mill with a 1 mm sieve, and collected in polyethylene vials for further laboratory analyses.

**In vitro rumen fermentation...**

Table 1. Diets ingredients, expressed in g.kg<sup>-1</sup> dry matter (DM)

	Oldman saltbush hay levels (% on DM)			
	8.4	18.8	31.2	48.3
<i>Ingredients (g.kg<sup>-1</sup> of dry matter)</i>				
Old man saltbush hay	84	188	312	483
Spineless cactus	749	576	370	72
Concentrate	167	236	318	445
R:C ratio	83.3:16.7	76.4:23.6	68.2:31.8	55.5:44.5
<i>Chemical composition of the diets (g.kg<sup>-1</sup> of dry matter)</i>				
Dry matter (%)	33,9	47,3	63,0	85,7
Organic matter	886	884	883	880
Mineral matter	114	116	117	120
Crude protein	125	137	135	138
Neutral detergent fiber	239	252	272	307
Acid detergent fiber	161	170	183	193
Lignin	39	46	53	63
Cellulose	122	124	130	130
Hemicellulose	78	82	89	114
Total carbohydrates	746	731	735	728
Non-fibrous carbohydrates	507	479	463	421
Ether extract	16	16	15	14
TDN	743	737	667	680
ME (kcal/kg of DM)	2687	2663	2411	2458

R:C - roughage:concentrate; \*ADIN - Acid detergent insoluble nitrogen; \*\*NDIN - Neutral detergent insoluble nitrogen; NDFp – neutral detergent fiber corrected for protein; ADFp – acid detergent fiber corrected for protein.

Analyses of the dry matter (DM), crude protein (CP) and mineral matter (MM) of the feeds (Table 2) and diets (Table 1) were performed in the Laboratories of Animal Nutrition of Semiarid

Embrapa, according to methodologies no. 7.007, 7.015 and 7.009 as described by the AOAC (Official..., 1984).

Table 2. Chemical composition of feeds, expressed in g.kg<sup>-1</sup> dry matter (DM)

Variables	Forage cactus	Oldman saltbush	Concentrates of the treatments			
			8.4	18.8	31.2	48.3
Dry matter (%)	14.6	91.4	91.5	92.0	91.4	91.0
Organic matter	882	847	924	919	919	916
Mineral matter	118	153	76	81	81	84
Crude protein	45	88	498	402	286	208
ADIN*	1	2	5	4	6	3
NDIN**	5	4	11	11	16	8
NDFp	238	516	104	75	73	91
ADFp	170	346	30	32	38	30
Lignin	37	114	11	13	13	11
Cellulose	133	232	19	19	25	19
Hemicellulose	68	170	74	43	35	61
Ether extract	17	10	13	16	16	18
Total carbohydrates	819	749	416	499	624	690
Non-fibrous carbohydrates	581	233	312	424	551	599

\*ADIN - Acid detergent insoluble nitrogen; \*\*NDIN - Neutral detergent insoluble nitrogen; NDFp – neutral detergent fiber corrected for protein; ADFp – acid detergent fiber corrected for protein.

The contents of total carbohydrates (TCHO) were calculated using the equations described previously by Sniffen *et al.* (1992). Concentrations of neutral detergent fiber (NDF, with the addition of thermostable amylase), acid detergent fiber (ADF, determined sequentially due to the presence of soluble carbohydrates) and lignin (LIG, in 72% sulfuric acid) were determined according to methods described previously by Van Soest *et al.* (1991), with further correction for the protein present in NDF and ADF, and the levels of non-fibrous carbohydrates (NFC), cellulose (CEL) and hemicelluloses (HEM) were estimated using those authors equations described previously. The neutral detergent-insoluble nitrogen (NDIN) and acid detergent-insoluble nitrogen (ADIN) contents were assessed using techniques described previously by Licitra *et al.* (1996).

For the energetic values of the diets, the data obtained from an experiment assessing apparent digestibility were applied to the following equation: TDN (%) = DNFC + DCP + (DEE × 2.25) + DNDF, in which “D” corresponds to the apparent digestibility of each referenced component (National..., 2001). The metabolizable energy was determined as described in a report by Harris (1970), which assumed that TDN = 4.409 × DE (digestible energy) and metabolizable energy (ME) = 0.82 × DE.

The evaluation of the rumen fermentation kinetics of the diets and feeds was performed by the semi-automated *in vitro* gas production technique described previously by Maurício *et al.* (2003) at the laboratory of gas production of Semiárid Embrapa.

The feeds containing varying proportions of oldman saltbush were weighed separately and then were mixed. One gram of each mixed-feed diet was added into individual fermentation vials

(160 mL) that were previously injected with CO<sub>2</sub>.

The ruminal fluid used as inoculum was obtained individually from three adult castrated male Sindi cattle via ruminal cannula. The animals were maintained for three days prior to collection on a diet consisting of oldman saltbush and forage cactus *ad libitum*, without defined proportions, plus 1kg.day<sup>-1</sup> of feed concentrate.

The ruminal fluid of each animal was filtered separately through a nylon sieve and maintained under continuous injection of CO<sub>2</sub> in thermal bottles that were previously heated to 39°C. Three vials were utilized per sample for the fluid from each of the animals (totaling 9 samples per diet or feed assessed). Ten milliliters of ruminal fluid were inoculated in each of these vials, which were then sealed with rubber corks (14 mm), placed in a polystyrene container, manually shaken (every hour), and kept in a temperature-controlled room at 39 °C.

Vials containing only ruminal fluid and culture medium (buffer) were used as blanks.

The pressure originating from the gases that accumulated within the upper part of the vials was measured using a pressure transducer (PressDATA 800) connected to a needle (0.6 mm). Readings were taken at 2, 4, 6, 8, 10, 12, 14, 17, 20, 24, 28, 34, 48, 72, and 96 h. The pressure data (P in psi - pressure per square inch) were converted into measurements of gas volume (V) by applying the quadratic equation:  $V = 0.17454 P^2$  (s.e. 0.0916) + 4.09089 P (s.e. 0.0637) + 0.00315 (s.e. 0.003), R<sup>2</sup> = 0.99.

The data from the cumulative production of the gases were analyzed by the two-compartmental model described previously by Schofield *et al.* (1994):

$$V(t) = \frac{Vf1}{[1 + e^{(2-4m1(Lag-T))}]} + \frac{Vf2}{[1 + e^{(2-4m2(Lag-T))}]}$$

In this equation,  $V(t)$  represents the total maximal volume of gases produced;  $Vf1$  represents the maximal volume of gas for the rapidly digested fraction (NFC);  $Vf2$  represents the maximal volume of gas for the slowly digested fraction (FC);  $m1$  stands for the specific growth rate for

the rapidly degraded fraction;  $m2$  stands for the specific growth rate for the slowly degraded fraction;  $Lag$  represents the duration of the initial events of digestion (lag period), which is present in the two phases; and  $T$  stands for the time of fermentation.

### In vitro rumen fermentation...

The fermentation residues were obtained by filtration of the contents of the fermentation vials through filter crucibles (porosity 1). These residues were dried at 105 °C until a constant weight was reached, and the results were used for the calculations of *in vitro* DM degradability.

For the evaluation of degradation kinetics (gravimetric technique), the exponential decay model, corrected for the lag period, was applied as described previously by Snedecor & Cochran (1989):

$$Y = b' - B * \exp(-c*t),$$

In this equation,  $Y$  represents the residue of DM at time  $t$ ;  $b'$  represents the potential degradation of the DM fraction;  $B$  represents the potentially degradable insoluble fraction, which will become degradable as a function of time at degradation rate  $c$ ;  $\exp$  represents the base of the natural logarithms;  $c$  represents the degradation rate of fraction  $B$  per time unit ( $\text{h}^{-1}$ ); and  $t$  represents the incubation time. The Parameters of rumen fermentation range and kinetics of the feeds obtained are demonstrated in Table 3.

Table 3. Parameters of rumen fermentation range and kinetics of the feeds

Variables	Forage cactus	Oldman saltbush	Concentrates			
			8.4	18.8	31.2	48.3
vNFC (mL)	222.5	76.1	133	183.3	197.2	208.8
kNFC ( $\text{h}^{-1}$ )	0.082	0.11	0.117	0.111	0.088	0.107
vFC (mL)	105.5	47.8	133.4	138.6	138.5	144.4
kFC ( $\text{h}^{-1}$ )	0.022	0.036	0.033	0.028	0.025	0.023
Lag (h:min)	01h:38min	04h:15min	05h:03min	04h:54min	04h:03min	04h:59min

vNFC - volume of gases from non-fibrous carbohydrates; kNFC - production rate of gases from non-fibrous carbohydrates; Lag - lag phase; vFC - volume of gases from fibrous carbohydrates; kFC production rate of gases from fibrous carbohydrates.

The data were subjected to variance analysis to determine the effects of different levels of oldman saltbush hay. Regression analysis was utilized for the significant effects ( $P < 0.05$ ) of the model using the software Sistema de Análises Estatísticas e Genéticas (Sistema..., 2007).

### RESULTS

The addition of oldman saltbush to the diets resulted in a quadratic effect in the volume of gases that originated from the NFCs. The diet containing 8.4% oldman saltbush yielded greater gas production and, consequently, more fermentation (Table 4).

Table 4. Volume of gases from non-fibrous carbohydrates (vNFC), production rate of non-fibrous carbohydrates (kNFC), lag phase (Lag), volume of gases from fibrous carbohydrates (vFC) and production rate of gases from fibrous carbohydrates (kFC) of experimental diets

Parameters	Oldman saltbush hay levels (%)				L	Q	Mean	CV
	8.4	18.8	31.2	48.3				
vNFC (mL)	185.0	153.8	141.7	143.0	0.020	0.030 <sup>1</sup>	155.9	12.64
kNFC ( $\text{h}^{-1}$ )	0.092	0.096	0.090	0.081	ns	Ns	0.090	11.16
vFC (mL)	111.7	113.3	107.4	114.0	ns	Ns	111.6	22.50
kFC ( $\text{h}^{-1}$ )	0.028	0.030	0.029	0.026	ns	Ns	0.028	15.92
Lag (h:min)	1h:24min	1h:13min	1h:57min	3h:41min	0.001	<0.0001 <sup>2</sup>	2:04	17.76

L - linear; Q - quadratic; CV - coefficient of variation; ns - not significant. <sup>1</sup> $\hat{Y} = 231.0833 - 54.4733x + 8.1333x^2$  ( $r^2 = 0.99$ ); <sup>2</sup> $\hat{Y} = 2.573 - 1.64536x + 0.48109x^2$  ( $R^2 = 0.99$ ).

The gas production rates from the NFCs of the diets did not differ and exhibited an average of  $0.090 \text{ h}^{-1}$ . The addition of oldman saltbush hay into the diets decreased the gas production, with the lowest gas volume of 136.48 mL resulting from the addition of 38.2% hay. The lag time varied quadratically; the shortest time resulted from the addition of 17.25% oldman saltbush hay into the diet, from which there was an increase during this period.

The volume and production rate of gases from the fibrous carbohydrates (FCs) were not affected by the levels of oldman saltbush hay added to the diets and exhibited average values of 111.6 mL and  $0.28 \text{ h}^{-1}$ , respectively. The maximum potential of gas production was observed in the diet containing 8.4% oldman saltbush hay and 74.9% forage cactus, which

yielded a gas volume of 296.7 mL produced due to degradation of the FC and NFC fractions ( $v_{FC} + v_{NFC}$ ).

A significant decrease in gas production was observed in response to the altered levels of NFCs resulting from the addition of oldman saltbush hay to the diets. There was no significant difference in the potential degradation of DM and OM, which exhibited mean values of 85.8% and 90.9%, respectively. However, the potentially degradable insoluble DM fractions exhibited quadratic behavior, which was proportional to the amounts of oldman saltbush hay added to the diets. The diet containing 8.4% exhibited the lowest percentage of the potentially degradable insoluble fraction (B) and the highest degradation rate of this fraction (Table 5).

Table 5. Means of the parameters of *in vitro* degradation kinetics of dry matter (DM) and organic matter (OM), in function of oldman saltbush hay levels in the diets

Parameters	Oldman saltbush hay levels (%)				L	Q	Mean	CV
	8.4	18.8	31.2	48.3				
DM b (%)	86.2	85.9	87.0	83.9	ns	ns	85.8	1.5
DM B (%)	65.4	74.6	75.5	80.3	*	* <sup>1</sup>	73.9	6.9
DM Kd ( $\text{h}^{-1}$ )	0.07	0.06	0.06	0.06	* <sup>2</sup>	ns	0.06	9.1
OM b (%)	91.6	91.1	92.6	88.4	ns	ns	90.9	2.0
OM B (%)	71.1	79.5	79.1	81.9	* <sup>3</sup>	ns	77.9	6.2
OM Kd ( $\text{h}^{-1}$ )	0.09	0.09	0.08	0.08	*	* <sup>4</sup>	0.08	8.2

b - potential degradation; B - potentially degradable insoluble fraction; Kd - degradation rate of fraction B; <sup>1</sup> $\hat{Y}=57.059 + 10.028x - 1.093x^2$  ( $R^2=0.93$ ); <sup>2</sup> $\hat{Y}=0.070 - 0.004x$  ( $R^2=0.99$ ); <sup>3</sup> $\hat{Y}=69.929 + 3.188x$  ( $R^2=0.77$ ); <sup>4</sup> $\hat{Y}=0.099 - 0.0092x + 0.00076x^2$  ( $R^2=0.97$ ); ns - not significant.

## DISCUSSION

The forage sources in the first diet (8.4% of oldman saltbush hay) were largely derived from forage cactus (74.9%), and due to the composition of this cactus (high NFC content and reduced NDF and LIG content) and the gas production observed during its fermentation (Table 3), it is possible that these factors contributed to the greater volume of gases that was produced by this diet.

Oldman saltbush grass is rich in lignins ( $113 \text{ g.kg}^{-1}$  in DM), and the susceptibility of its different carbohydrates to bacterial degradation varies greatly according to their physicochemical properties or other factors that limit the access of bacterial enzymes to their substrates. Thus, the decreased gas production might directly reflect

the increased proportions of oldman saltbush hay in the diets.

The addition of hay levels in excess of 38.2% promoted increases in the gas production from NFCs. This observation might be attributed to the fermentative characteristics of the concentrate, which was present at 44.5% in the diet with 48.3% hay, rather than to the levels of hay, which also contributed to the volume of gases produced by the NFCs (208.8 mL) of concentrate 4 (Table 3).

In contrast, Moreira *et al.* (2010) reported that increased gas production was associated with the silages of sorghum and corn. The sources of carbohydrates (flint corn, dent corn and citrus pulp) exhibited higher fermentation rates, which possibly result from the increased production of acetate from the fermentation of the FCs present

in the silages effects that were contrary to those observed in this study.

Fermentation gases are produced predominantly when the substrates are fermented to acetate and butyrate; lower amounts of gases are associated with the reaction of propionate with the culture medium (indirect production). Soluble carbohydrates produce relatively higher amounts of propionate, whereas the inverse is observed with FCs (El-Waziry, 2007). However, because of the greater availability of rapidly fermentable carbohydrates, more gas is produced from the NFCs in feeds that contain this nutrient, a notion that is consistent with our findings.

The shorter lag time (Lag) observed might be attributed to the physicochemical characteristics of the forage cactus, which was present in significant amounts (576 g.kg<sup>-1</sup> in DM) in the diet composed of 18.8% hay. The soluble fraction constitutes an energetic substrate of rapid fermentation that facilitates the adhesion and colonization of microorganisms, consequently elevating the fermentation of FC and reducing the Lag period. However, the importance of the soluble fraction starts to decrease when greater amounts of the components of the cell wall become dispensable (Noguera *et al.*, 2005).

According to Detman *et al.* (2009), although the lag represented by the model of Schofield *et al.* (1994) results from mixed origins (i.e., common to the two digestion compartments), the fibrous fraction can be directly associated with most of the lag-related events, thereby accounting for the greater lag observed with feeds containing higher proportions of lignin. The diet composed of 48.3% oldman saltbush hay exhibited a greater Lag period (3 hours and 41 minutes). The longer Lag period observed in the diet with 48.3% hay might be attributed to the proportions of NDF (331 g.kg<sup>-1</sup> DM) and LIG (63 g.kg<sup>-1</sup> DM) in this diet.

However, high fermentation potential might be related to the profile of volatile fatty acids produced. The forage cactus contains high levels of pectin (Batista *et al.*, 2003), which contributes to a greater production of acetate. Lower gas production is observed when propionate is

formed, due to the absence of CO<sub>2</sub> production. However, when fermentation is directed toward the formation of acetate and butyrate, the CO<sub>2</sub> is produced, consequently increasing the volume of gases. In such cases the fermentation of the forage cactus results in the production of gases derived from NFCs, due to the fermentative property of pectin. The greater levels of pectin in the cactus-containing diets are reflected in the rate of gas production derived from NFCs (Figure 1).

The cumulative gas production and the contributions of the NFC and FC fractions in the four diets evaluated suggest that the fibrous fraction is more extensively degraded in the diet containing 8.4% hay (Figure 1), possibly due to the carbohydrate profile (i.e., pectin content) present in the cactus that renders it more easily degraded by the ruminal microorganisms. According to Van Soest (1994), the faster degradation of pectin for insoluble fibrous compounds results in greater production of acetate and fermentation gases.

These results might be associated with the roughage:concentrate ratio of the diets. Higher proportions of concentrate increase the availability of protein and carbohydrates in a synchronous manner, which might alter the acetate:propionate ratios, leading to greater efficiency of microbial degradation and usage of dietary N. Thus, although lower production of gases (from NFC and FC) was observed, the efficiency of diets with greater content of oldman saltbush was higher.

Lower production of gases attributed to increased FC and LIG content has been reported previously. Pereira *et al.* (2005) evaluated sunflower silages and observed reduced gas production from the more mature silages and silages with higher FC content. According to Jung and Allen (1995), the diversity of cell wall constituents is associated with variation in the digestibility of nutrients, which can be intrinsically linked to morphogenetic characteristics of the plants. Lignin is a phenyl propanol polymer that is found in the cell wall, and according to these authors, lignin composition and concentration might affect the rate and extent of fiber digestion.

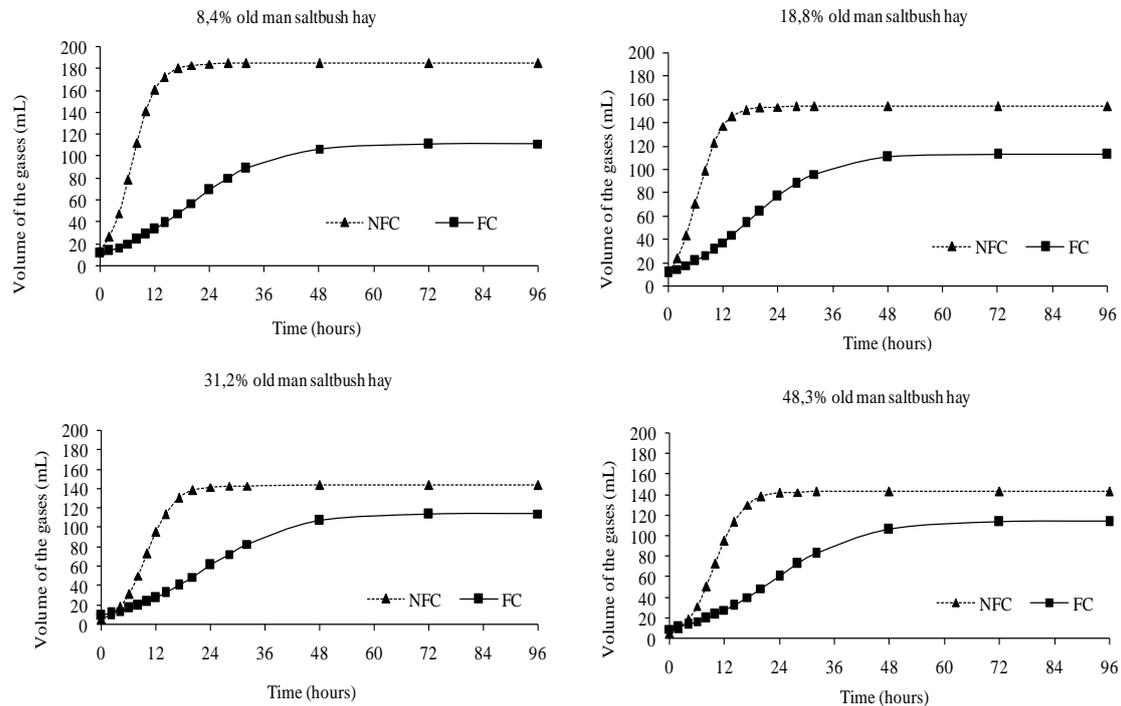


Figure 1. Gas production from the non-fibrous carbohydrates (NFC) and fibrous-carbohydrates (FC), due to oldman saltbush hay levels in the diets.

The possible low pH due to the elevated levels of NFCs in this diet might reduce the degradation-resistant insoluble fraction (B). The insoluble DM fraction is associated with the fibrous fraction; thus, one can infer that the lower B in this diet is attributed to the following two effects related to the presence of NFC:pH effect and concentrate:carbohydrate effect (Mould *et al.*, 1983). Soluble carbohydrates and starch can block the digestion of cellulose by reducing ruminal pH by promoting competition between cellulolytic and amyolytic bacteria for essential nutrients that are distinct in energy content, or by promoting the usage of alternative energy sources by certain cellulolytic bacteria.

Gonçalves *et al.* (2001) compared the degradation kinetics of roughages utilized in the feeding of dairy goats when the roughages were subjected to different pH levels. The authors observed a decrease in the degradability of DM and NDF at lower pH values. Likewise, Costa *et al.* (2009) evaluated the *in vitro* degradation of elephant grass supplemented with different sources of carbohydrate and proteins, demonstrating a decrease in the degradation rate of NDF that was attributed to increased amounts

of soluble carbohydrates. Costa *et al.* (2008) also found a decrease in the degradation of fiber from low-quality tropical forages that were supplemented with starch. However, the addition of pectin did not impair the degradation of potentially degradable NDF. The authors attributed these results (lower degradation of fiber and DM) to the competition for nutrients between the ruminal microorganisms, which preferentially utilize soluble carbohydrates rather than FCs as energy sources.

Thus, the increased degradation rates of the fraction of DM and OM (Table 6) exhibited by the diet with 8.4% hay and 74.9% cactus were possibly attributed to the availability of carbohydrates that supplied the energy to increase the proliferation of the ruminal microorganisms.

The increased potentially soluble fraction of DM and OM and reduced degradation rates of this fraction (Table 5) due to the addition of hay in the diets can be associated with the physicochemical characteristics of the hay, which has an elevated lignin content ( $113 \text{ g.kg}^{-1}$  DM).

## CONCLUSIONS

The addition of 8.4% hay in the diets resulted in the maximal gas production from the fibrous fraction of diets containing cactus and oldman saltbush hay. The combination of oldman saltbush hay with forage cactus represents an option for the roughage supplementation of cattlefeeds in semi-arid regions.

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