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Effects of a *Saccharomyces cerevisiae* yeast on ruminal fermentation and fibre degradation of maize silages in cows☆

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Abstract

Effects of a *Saccaromyces cerevisiae* yeast (Levucell SC 10 ME; 1×10^{10} CFU/g (SC)) on ruminal fermentation and fibre degradation of maize silages was studied with 3 non-lactating fistulated cows fed maize silage, concentrate and meadow hay (48:42:10, DM basis) twice daily and supplemented with 0 (SC0), 0.3 (SC0.3) and 1 g (SC1.0) of SC/day. Maize silages, 40, were ruminally incubated *in situ* for 36 h to determine neutral detergent fibre degradation (NDFdeg). Silages were divided into two groups according to NDFdeg measured with the SC0 diet, being a low fibre degradation (LFD) group (NDFdeg: 0.20–0.30) and a high fibre degradation (HFD) group (NDFdeg: 0.35–0.45). Rumen fluid was collected on 2 non-consecutive days at 0, 2, 4 and 8 h post-feeding for determination of pH, ammonia N, volatile fatty acids (VFA) and lactate concentrations. The study was a 3×4 factorial design, with 3 replications, to examine effects on ruminal fermentation and in a 3×2 factorial design,

Abbreviations: ADFom, acid detergent fibre expressed exclusive of residue ash; aNDFom, neutral detergent fibre assayed with heat stable amylase; CP, crude protein; DM, dry matter; EE, ether extract; HFD, high fibre degradation group; LFD, low fibre degradation group; Lignin(sa), sulphuric acid lignin; NDFdeg, rumen degradation of NDF after 36 h of ruminal *in sacco* incubation; NDFom, neutral detergent fibre expressed exclusive of residue ash; NFC, non-fibre carbohydrates; SC, *Saccharomyces cerevisiae*; SC0, basal diet without *Saccharomyces cerevisiae*; SC0.3, basal diet with 0.3 g *Saccharomyces cerevisiae*/day; SC1.0, basal diet with 1 g *Saccharomyces cerevisiae*/day; VFA, volatile fatty acids.

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with 3 replications, to examine effects on NDFdeg. Inclusion of SC increased (P<0.01) ruminal pH, decreased (P<0.01) lactate concentration and the acetate:propionate (P<0.01) ratio, but had no effect on ammonia N concentration. The SC addition at 1 g/day increased VFA concentration *versus* the control diet (P<0.01), further the reduced acetate:propionate ratio and increased fibrolytic activity of rumen bacteria as assessed by NDFdeg of silages. No effect on silage degradation occurred with the SC0.3 diet. Changes of rumen fermentation occurred from 0 to 8 h post-feeding, as expected, increasing (P<0.05) from 0 to 2 and/or 4 h after feeding followed by a decrease to 8 h. There was only an interaction (*i.e.,* P<0.05) of diet × time post-feeding for ruminal pH and lactate concentration. Results show that this SC strain was effective in alleviating pH depression and lactate concentration after feeding of fistulated cows fed twice daily at close to the metabolizable energy maintenance requirements, irrespective of the level of YC inclusion. This suggests that this SC has the potential to reduce the risk of rumen acidosis in commercial cattle fed maize silage based diets and, if used at the highest level, could be of further benefit due to increased fibre degradation of low quality maize silages.

Keywords: *Saccharomyces cerevisiae*; Ruminal fermentation; *In sacco* fibre degradation; Cows

1. Introduction

Research to improve rumen function to increase production efficiency of domestic ruminants has been a priority of nutritionists for many years, in order to reduce energy and nutrient losses from commercial ruminant production. Antibiotics included in diets in subtherapeutic levels have proven to be effective tools for this purpose (McDonald et al., 2002), but during the last two decades there has been a growing concern regarding use of these substances in animal feed industry leading to a ban on their utilization in the EU. This has shifted the focus of researchers to the study, and utilization, of non-antibiotic alternatives to manipulate rumen fermentation in order to reduce energy and nutrient losses from commercial ruminant production systems.

Among these alternatives, viable and non-viable yeast cultures, mainly from strains of *Saccharomyces cerevisiae* (SC), have been the most studied, and used, in dairy and beef cattle operations. Robinson (2002) summarized studies that have been published in the scientific literature between 1981 and 2002 to examine impacts of specific commercial yeast products, and concluded that most published studies showed consistent, although small (*i.e.,* 2.0 to 3.7%), improvements in dry matter (DM) intake and production.

Cattle responses attributed to yeast are usually related to stimulation of cellulolytic bacteria (Newbold et al., 1996) thereby increasing the potential to enhance fibre digestion in the rumen and to its ability to prevent a decline in rumen pH by decreasing lactic acid production and/or increasing utilization of lactic acid by some bacteria (Chaucheyras et al., 1996). From the review (Robinson, 2002), it appears that animal responses to yeast products are independent of the diet. However, experimental support for uniform effects of SC on rumen fibre degradation, regardless of the digestibility of the basal diet is inconsistent (Roa et al., 1997; Krehbiel et al., 2006).

This experiment was designed with rumen-fistulated non-lactating cows fed maize silage as the main forage to examine effects of a commercial strain of SC on *in situ* ruminal degradation of 40 samples of maize silage that differed in chemical composition and *in situ*

Table 1
Dry matter (DM; g/kg) and chemical composition[a] (g/kg DM) of feeds and diet fed (±S.D.)

	Maize silage	Meadow hay	Concentrate[b]	Diet
DM	331 ± 9.0	898 ± 12.4	905 ± 4.2	663
Chemical composition[c]				
Ash	45 ± 2.8	63 ± 3.3	92 ± 1.6	67
CP	70 ± 2.8	76 ± 1.8	222 ± 4.6	134
EE	27 ± 1.6	10 ± 1.2	38 ± 1.0	30
aNDFom[d]	447 ± 5.9	641 ± 15.4	227 ± 5.2	374
ADFom	252 ± 8.7	399 ± 11.0	119 ± 2.2	202
Lignin(sa)	18 ± 5.4	55 ± 6.3	33 ± 1.9	29
Starch	281 ± 2.2	ND	208 ± 4.1	222

ND, not determined.

[a] Each value is the mean of 9 samples of maize silage and 6 samples of meadow hay and concentrate.

[b] Composition (kg/tonnes): barley grain, 100.0; maize grain, 88.4; wheat grain, 125.0; liquid molasses, 25.0; calcium soaps, 7.5; beet pulp, 45.0; corn gluten feed, 34.8; dried distillers maize grains, 204.9; sunflower meal, 96.9; soybean meal, 176.8; full fat soybeans, 45.0; sodium bicarbonate, 8.5; magnesium oxide, 6.5; calcium carbonate, 18.7; common salt, 5.0; mineral and vitamin premix, 2.0; binder, 10.0.

[c] CP: crude protein; EE: ether extract; aNDFom: neutral detergent fibre assayed with heat stable amylase; ADFom: acid detergent fibre expressed exclusive of residue ash; lignin(sa): sulphuric acid lignin.

[d] For meadow hay, NDFom was assayed without heat stable amylase.

ruminal degradation. Ruminal activity was also assessed through pH, volatile fatty acids (VFA), lactate and ammonia N measurements.

2. Materials and methods

2.1. Animals, diets and experimental design

Three non-lactating cows with a live weight of 538 ± 12.6 kg fitted with a rumen cannula were fed twice daily at 08:00 and 16:00 h at 1.20 times maintenance level of metabolizable energy (AFRC, 1993) with a complete diet consisting of maize silage, a commercial concentrate and meadow hay (48:42:10, DM basis). The chemical composition of feeds and diet is in Table 1. The three dietary treatments were the basal diet without SC supplementation (SC0), the basal diet supplemented with 0.3 g of SC/day (SC0.3), and the basal diet supplemented with 1 g of SC/day (SC1.0). According to the manufacturer (Lallemand, Vienna, Austria) of the SC (Levucell SC 10 ME), the number of CFU was 1×10^{10}/g (lot number 221317v519;). The SC0.3 treatment was included since the recommended level of this SC formulation is 1 g/day for lactating dairy cows according to the manufacturer and the DM intake of cows used in this study was approximately 0.30 of the DM intake of a high producing dairy cow at peak lactation. Fresh water was available at all times *ad libitum*, and cows were housed in individual tie stalls throughout the experiment.

The SC was offered as a powder obtained by freeze drying and it dosed directly into the rumen. Ground (1 mm) concentrate (50 g) was used as a carrier and mixed with the SC before it was dosed to the rumen. The mixture (SC and carrier) was introduced through the rumen cannula daily just before the morning feeding. To eliminate any carrier effect, 50 g of ground concentrate was also dosed into the rumen when animals were fed the SC0 diet.

Each experimental period lasted approximately 30 days and was comprised of 10 days for dietary adaptation, 15–18 days for the degradation study described below and 2 days for rumen liquor sampling. This study was preceded by a pre-experimental period without SC addition (*i.e.,* diet SC0) for selection of maize silage samples to be used. When there were no *in sacco* bags in the rumen, cows were allowed to exercise outside for 3 h/day.

Samples of the feeds offered to the cows were collected weekly and bulked after each experimental period. Bulked samples were dried at 60 °C for at least 24 h in a forced-air oven for DM determination. Dry samples were prepared for chemical analysis by grinding through a 1 mm screen (Retsch, SKM100, Haan, Germany).

2.2. Maize silages

Maize silages, 89, were collected from dairy farms in Northwest Portugal from unknown varieties of maize, but were all grown and ensiled in 2005. Samples of silages were dried in a forced-air oven at 65 °C for 48 h and ground through a 4-mm screen before use.

All silages were incubated *in situ* for 36 h to determine neutral detergent fibre degradation (NDFdeg) using the SC0 diet. Then, 49 samples were selected and divided into two groups according to its NDFdeg (*i.e.,* low fibre degradation (LFD) group ($n = 20$) with NDFdeg varying between 0.20 and 0.30 and high fibre degradation (HFD) group ($n = 20$) with NDFdeg varying between 0.35 and 0.45).

2.3. Rumen fluid collection

On 2 non-consecutive days during the incubation period, approximately 50 ml of rumen fluid were collected from the middle of the rumen of each cow at 0 h (before feeding) and 2, 4 and 8 h after the morning feeding. The pH values of the rumen fluid were measured immediately with a digital pH meter (Wissenschaftlich-Technische Werkstätten, pH 530, Weilheim, Germany). Thereafter, samples were kept frozen at −20 °C for ammonia N, VFA and lactate assay.

2.4. Degradation study

In sacco nylon bags (Nybolt PA 40/30, Zurich, Switzerland) with approximately 5 g of DM (4 mm grind) samples were ruminally incubated according to the method of Ørskov et al. (1980) but following procedures of Guedes and Dias-da-Silva (1994). Bags were put into the rumen of each cow 2 h after the morning feeding and withdrawn after 36 h. Incubations were repeated once in each cow making, in total, 6 replications/sample. After removal from the rumen, bags were washed with tap water in a washing machine (TS803, Balay, Zaragoza, Spain), and dried at 65 °C for at least 24 h in a forced-air oven. Bags were weighed and residues analysed for neutral detergent fibre (NDF). Neutral detergent fibre degradation (NDFdeg) was expressed as a coefficient.

2.5. Laboratory analyses

Dry and ground (1 mm) feeds and maize silage samples were analyzed for ash (method no 942.05) and ether extract (EE; method no. 92039) of AOAC (1990). The N was assayed

by a Kjeldahl method (AOAC, 1990; method no. 954.13 988.05), modified by using a solution of boric acid (40 g/l) to receive free ammonia during distillation, a solution of 2 g/l of bromocresol green and 1 g/l of methyl red in ethanol was used as indicators and a standard acid solution (1N HCl) was used for titration. Starch was measured by the method proposed by Salomonsson et al. (1984) using a spectrometer (U2000, Hitachi Ltd., Japan).

Neutral detergent fibre (NDFom) was expressed on an ash-free basis (Van Soest et al., 1991) and sodium sulphite was not used. For concentrate and maize silage samples, α-amylase was used prior NDFom analysis (aNDFom). Acid detergent fibre (ADFom) is expressed without residual ash (AOAC, 1990, method no. 973.18). Sulphuric acid lignin (lignin(sa)) was analysed according to Robertson and Van Soest (1981). Neutral detergent fibre expressed with residual ash was used for the determination of NDFdeg.

Samples of rumen fluid were centrifuged at 10,000 × *g* for 15 min at 4 °C and supernatants were collected for ammonia N, VFA and lactate analysis. Ammonia N was determined using an automated analyser according to Novozamsky et al. (1974). Concentrations of VFA were determined by gas–liquid chromatography (Shimadzu GC-141 B, Kyoto, Japan) using pivalic acid as internal standard according to Czerkawski (1976). Separation of acetate, propionate and butyrate used a capillary column (Supelco Nukol, 0.25 mm i.d. × 30 m × 0.25 μm), operated at 135 °C using helium as the carrier gas. Injection block temperature was 210 °C. Quantification of acids used a flame ionization detector at 180 °C connected to an integrator (Shimadzu C-R6A Chromatopac, Kyoto, Japan). Lactate was determined using an enzymatic assay procedure (K-DLate 02/06, Megazyme, Ireland).

2.6. Statistical analyses

Statistical analyses used SAS (1990) software. Fermentation data (*i.e.,* pH, NH_3N, acetate, propionate, butyrate, total VFA, lactate, acetate:propionate) were analysed using repeated measures data of MIXED procedure with the model:

$$Y_{ijk} = \mu + D_i + T_j + (DT)_{ij} + A_{ik} + \varepsilon_{ijk}$$

where μ is the overall mean, D_i the fixed effect of diet ($i = 1$–3), T_j the fixed effect of time after feeding ($j = 1$–4), $(DT)_{ij}$ the fixed interaction effect of D_j and T_j, A_{ik} is the random effect of animal within D_i, and ε_{ijk} is the random error. Time after feeding was used as repeated measure. Statistical analysis of NDFdeg was carried out using the GLM procedure of SAS (1990) according to a 3 × 2 factorial design with SC level and maize silage quality as factors. Differences between treatment means were determined by Student's multiple range *t*-test.

3. Results

3.1. Chemical composition of maize silages

Means, minimum, maximum and standard deviation (S.D.) of the chemical composition of maize silages are in Table 2. Most chemical components showed a wide range. The highest variation was for starch content (*i.e.,* 251 ± 45.6) with lower variation for CP (*i.e.,* 80.8 ± 6.63). The aNDFom represented the highest proportion of maize silage DM, being 479 ± 47.3 g/kg.

Table 2
Mean minimum and maximum values and standard deviation (S.D.) of the dry matter (DM; g/kg) chemical composition (g/kg DM) of maize silages ($n = 40$)

	Mean	Minimum	Maximum	S.D.
DM	326	270	429	38.6
Chemical composition[a]				
Ash	44.8	38.5	61.1	5.51
CP	80.8	70.0	95.5	6.63
EE	28.7	21.3	34.3	3.40
aNDFom	479	385	582	47.3
ADFom	266	211	306	24.0
Lignin(sa)	31.4	21.6	37.8	3.92
Starch	251	170	378	45.6

[a] CP: crude protein; EE: ether extract; aNDFom: neutral detergent fibre assayed with heat stable amylase; ADFom: acid detergent fibre expressed exclusive of residue ash; lignin(sa): sulphuric acid lignin.

3.2. Ruminal fermentation

Ruminal pH was affected (P<0.01) by SC supplementation (Table 3). The pH values in SC0.3 and SC1.0 diets did not differ, but values tended (P=0.08) to be higher with the SC1.0 diet. Time after feeding affected (P<0.001) ruminal pH, with higher (P<0.05) values just before feeding (0 h) and the lower (P<0.05) values 2 and 4 h after feeding. Diet supplementation with SC reduced pH variation, which may explain the treatment × time interaction (P<0.05). The pattern of diurnal fluctuation of pH values and lactate concentration of ruminal fluid in cows is in Fig. 1, where each point is the mean of values of three cows.

Concentration of lactate was reduced (P<0.001) by SC supplementation, with higher (P<0.05) values for diet SC0. Rumen lactate concentration increased (P<0.05) from 0 to 2 h after the morning meal, but decreased (P<0.05) thereafter. There was an interaction

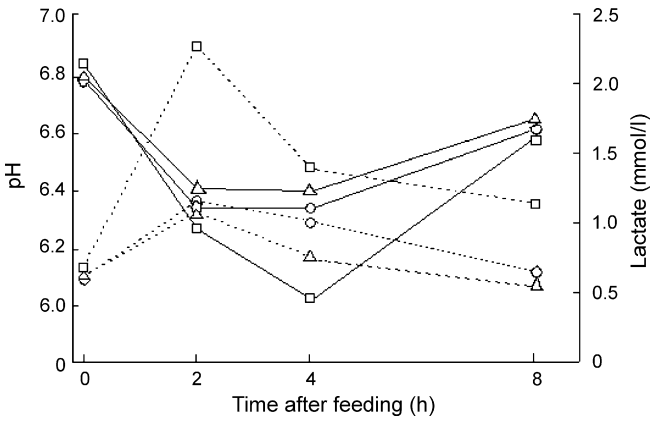

Fig. 1. Diurnal patterns of ruminal pH (—) and lactate concentration (- - -) in cows fed basal diet without *Saccharomyces cerevisiae* (SC; SC0; □), basal diet with 0.3 g SC/day (SC0.3; ◯) and basal diet with 1 g SC/day (SC1.0; △). Pooled standard error for pH: 0.036 (SCO), 0.041 (SC0.3) and 0.040 (SC1.0); pooled standard error for lactate: 0.026 (SCO), 0.030 (SC0.3) and 0.033 (SC1.0).

Table 3
Effects of *Saccharomyces cerevisiae* supplementation and sampling time on pH values and lactate (mmol/l), volatile fatty acids (VFA; mmol/l) and ammonia N (mg/l) concentrations in the rumen

Sampling time	Diets[a]												S.E.[b]	Probability		
	SC0				SC0.3				SC1.0					Diet (D)	Time (T)	DxT
	0h	2h	4h	8h	0h	2h	4h	8h	0h	2h	4h	8h				
pH	6.77 d	6.25 g	6.05 h	6.56[b]	6.77 d	6.34 fg	6.32 fg	6.59 e	6.78[a]	6.37[c] d	6.40[c]	6.63[ab]	0.039	0.0010	<0.0001	0.0350
Lactate	0.66 h	5.25 d	1.38 de	1.11 ef	0.59 h	1.13 ef	0.99 fg	0.62 h	0.60 h	1.05 f	0.74 gh	0.53 h	0.030	0.0004	<0.0001	0.0369
VFA																
Acetate	45.7	64.2	67.8	52.3	44.0	60.0	65.5	61.9	50.0	70.0	73.9	57.0	2.129	0.0389	<0.0001	0.205
Propionate	15.6	28.3	26.5	18.2	15.4	29.3	28.5	22.7	24.3	38.5	36.6	27.4	0.669	<0.0001	<0.0001	0.730
Butyrate	7.7	12.4	12.9	10.8	9.0	11.4	13.4	13.1	8.8	13.9	14.4	12.1	0.379	0.0452	<0.0001	0.321
Total	68.8	104.8	107.2	81.2	68.4	104.7	107.3	97.7	82.9	122.5	125.0	96.5	3.264	0.0006	<0.0001	0.243
A:P[c]	2.93 d	2.27 fg	2.56 e	2.87 de	2.86 d	2.05 fgh	2.30 f	2.73 de	2.06 fgh	1.82 h	2.02 gh	2.08 fgh	0.121	<0.0001	<0.0001	0.019
NH_3N	209	268	189	200	214	280	190	202	233	279	234	213	11.50	0.1441	<0.0001	0.944

Values in the same line with different letters (d–h) are differ according to the Student's *t*-test (P<0.05).

[a] SC0 basal diet without *Saccharomyces cerevisiae* (SC), SC0.3 basal diet with 0.3 g SC/day, SC1.0 basal diet with 1 g SC/day.

[b] Pooled standard error.

[c] Acetate:Propionate.

between diet and time after feeding (P<0.05) for lactate concentration in the rumen, with SC reducing (P<0.05) the extent of the increase of ruminal lactate concentration, which occurred 2 h after feeding and persisted for 4 h after feeding.

Ruminal ammonia N was affected (P<0.001) by sampling time with concentrations peaking (P<0.05) 2 h after feeding. The pattern of diurnal fluctuation of ruminal ammonia N was similar among diets.

Inclusion of SC in the diet increased acetate (P<0.05), propionate (P<0.001) and butyrate (P<0.05) concentrations in ruminal fluid. Acetate, propionate and butyrate concentrations were also affected (P<0.001) by sampling time with higher (P<0.05) values 2 and 4 h after feeding.

The acetate:propionate ratio was affected (P<0.001) by diet, increasing (P<0.05) from diet SC0 to diet SC0.3 and to diet SC1.0. Time after feeding also affected the acetate:propionate ratio, the lowest value (P<0.05) occurring 2 h after feeding. There was an interaction between diet and sampling time (P<0.05), reflecting a different diurnal pattern of acetate:propionate ratio among diets.

3.3. Neutral detergent fibre degradation of silage samples

An effect (P<0.01) of diet on degradation of NDF from maize silages samples suspended in nylon bags (NDFdeg) occurred (Table 4). While inclusion of 0.3 g/day of SC had no effect on NDFdeg, increasing the level to 1 g/day increased (P<0.05) NDFdeg. Effects of SC inclusion in the diet depended (P<0.05) on the initial NDF degradation of the silags. While SC addition at a level of 1 g/day increased (P<0.05) NDFdeg in the LFD group, it had no effect on HFD silages.

4. Discussion

4.1. Chemical composition of silages

The chemical composition of the maize silages are consistent with values for maize silages from the Northwest of Portugal (Dias-da-Silva and Lage, 2003). The wide range in starch content probably reflects large differences in their grain content. Generally, grain and DM content are positively correlated (Johnson et al., 1997; Fonseca et al., 2000), suggesting large variation in maturity of the silages used in our study as it is confirmed by the range of variation in DM content (*i.e.,* 270 to 429 g/kg). However, given the expected, but unknown, variability of the growing and harvesting conditions of the original maize crops, the genotypes used (Barrière et al., 1995) and silo management such as protection of silages from rain, the lack of a correlation between DM and starch content in the 40 samples was not unexpected.

4.2. Ruminal fermentation

It is well known that variation of ruminal pH during the day depends on many factors. Among the most important are the dietary content of rapidly fermentable non-fibre carbo-

Table 4
Effects of *Saccharomyces cerevisiae* supplementation and maize silage quality on neutral detergent fibre degradation (NDFdeg) of maize silages incubated in the rumen ($n = 40$)

	Diets[a]						S.E.[d]	Probability		
	SC0		SC0.3		SC1.0			Diet	Maize silage quality	Interaction
	LFD[b]	HFD[c]	LFD	HFD	LFD	HFD				
NDFdeg (units)	0.247 g	0.393 e	0.257 g	0.402 e	0.306 f	0.410 e	0.009	0.0009	<0.0001	0.0382

Values in the same column (e–g) within the same effect with different letters differ according to the Student's *t*-test (P<0.05).
[a] SC0 basal diet without *Saccharomyces cerevisiae* (SC), SC0.3 basal diet with 0.3 g SC/day, SC1.0 basal diet with 1 g SC/day.
[b] HFD group, high fibre degradability group.
[c] LFD group, low fibre degradability group.
[d] Pooled standard error.

hydrates (NFC), total daily intake of NFC, feeding diet components separately or mixed in a complete ration, frequency of ration feeding, animal and diet buffering capacity and adaptation of animals to diets high in NFC (Krause and Oetzel, 2006). In the present study, DM intake was restricted to approximately 1.20 times the maintenance requirements for metabolizable energy of the cows, feed was offered twice a day (08:00 and 16:00 h) and NFC density of the diet was judged to be moderate (Hall, 1999), which may explain why measured values of rumen pH in the control diet remained above 6.0. Thus it seems unlikely that the cows in the control treatment would have been at risk of acidosis. Under circumstances where pH of the control diet is 6.0 or higher, literature reports suggest little, or no, effect of SC supplementation on pH (Yoon and Stern, 1996; Doreau and Jouany, 1998; Enjalbert et al., 1999; García et al., 2000). However, in the present study, the strain of SC used, even at the lowest level of inclusion in the diet, was effective in alleviating pH depression that typically occurs after feeding.

It has been suggested that the elevating effect of SC on ruminal pH may be due to reduced lactate concentrations in the rumen (Williams et al., 1991), through the increase of activity of lactate-utilising bacteria such as *Selenomonas ruminantium* (Nisbet and Martin, 1991; Callaway and Martin, 1997) or *Megasphaera elsdenii* (Chaucheyras et al., 1996; Callaway and Martin, 1997) and/or the decrease of activity of lactate producing bacteria (Martin and Nisbet, 1992). Variations in pH observed in our study due to SC inclusion are consistent with the lactate concentrations (Fig. 1), where lactate concentrations in the SC0.3 and SC1.0 diets were only 39.5 and 34.8% of the control diet, respectively and appear to confirm results of the few reports where lactate concentrations have been measured either *in vivo* (Erasmus et al., 1992; Williams et al., 1991) or *in vitro* (Lila et al., 2004).

The increase in total VFA concentration in the rumen due to SC supplementation is not surprising, given the effects on pH and lactic acid above discussed, but this effect was much more pronounced with the highest dose of SC: 3.8 for SC0.3 *versus* 16.2 mmol/l for SC1.0 calculated over a period of 8 h from 4 sampling times. It is known that an increase in rumen pH favours fibrolytic activity of rumen bacteria (Mould et al., 1983–1984) which may explain, at least partially, the effect in this study. Most published results on effects of SC supplementation have shown that total VFA concentration was not affected (Dawson et al., 1990; Erasmus et al., 1992; Carro et al., 1992; Piva et al., 1993; Plata et al., 1994; Yoon and Stern, 1996; Doreau and Jouany, 1998; Enjalbert et al., 1999).

Besides the stimulating effect of SC on VFA concentrations, supplementation with SC also changed the molar proportion of VFA in the rumen to increase the glucogenic potential of the diet (*i.e.,* lower acetate:propionate ratio). Again, the highest level of SC inclusion was clearly more effective for this purpose, resulting in a higher increase in propionate concentration *versus* acetate. This suggests that effects of the SC studied go beyond lactic acid production and/or utilization, or improvement of fibre degradation. A lower acetate:propionate ratio due to SC supplementation also occurred in Williams et al. (1991) steers (forage to concentrate, 50:50) and by Erasmus et al. (1992) in lactating dairy cows (forage to concentrate, 43:57). In contrast, Mwenya et al. (2005) found an increase in this ratio when fistulated non-lactating cows were supplemented with *Trichosporum sericeum* yeast culture (forage to concentrate ratio, 70:30).

The lack of effect of SC on ammonia concentration in the rumen is consistent with previous results (Carro et al., 1992; Plata et al., 1994; Newbold et al., 1995; Yoon and

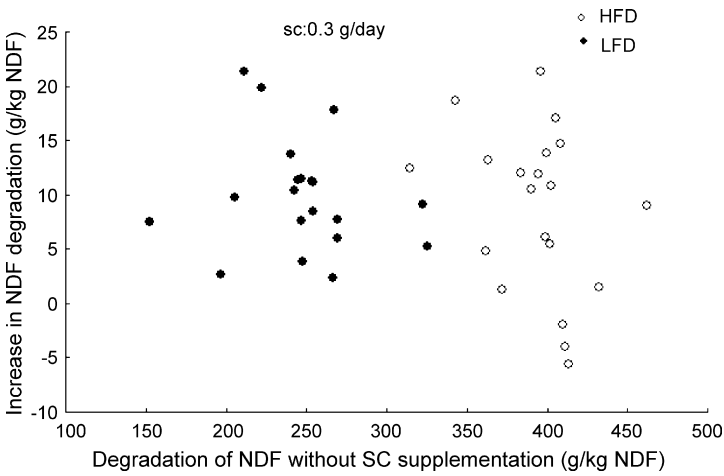

Fig. 2. Effects of supplementation with 0.3 g *Saccharomyces cerevisiae* per day (SC0.3) on fibre (NDF) degradation of maize silages after 36 h of incubation in the rumen of cows: HFD, high fibre degradation group and LFD, low fibre degradation group.

Stern, 1996; Doreau and Jouany, 1998), who found that it was not changed by addition of SC, and also to those by Mwenya et al. (2005) using *Trichosporum sericeum*. In contrast, decreased rumen ammonia N concentration with SC supplementation has been reported *in vivo* (Harrison et al., 1988; Erasmus et al., 1992; Piva et al., 1993; Enjalbert et al., 1999).

4.3. Neutral detergent fibre degradation

Effects of SC supplementation on ruminal fibre disappearance *in sacco* of our maize silages fully agree with the changes observed in rumen environment. In the 8 experiments reviewed by Robinson (2002) where fibre degradation was measured, a mean increase of 6.6% in NDF digestibility occurred after 24 h incubation *in sacco* in 75% of the experiments regardless of the feed incubated. The increase was lower when NDF was measured in the entire digestive tract (3.9%; 5 of 6 experiments) and not existent *in vitro* (−1.1%; 3 of 5 experiments). As noted above, fibrolytic activity was unchanged when the basal diet was supplemented with the low level of yeast (Fig. 2). When 1.0 g of SC was used, closer observation of the data showed that the increase in NDF disappearance from the bags was inversely related to the initial degradability of the silages (*i.e.,* degradability measured without yeast supplementation; Fig. 3), where the linear was:

$$\begin{aligned}&\text{Increase in NDFdeg(g/kg NDF)}\\&\quad = -0.2685\,\text{initial NDFdeg(g/kg NDF)}\\&\qquad + 123.975(n = 40; r^2 = 0.85; \text{R.S.D.} = 9.23; \text{P} < 0.001)\end{aligned}$$

The increase in NDFdeg as a result of 1.0 g inclusion of SC in the diet was 59 and 18 g/kg NDF in LFD and HFD groups of silages, respectively. Besides indicating that SC

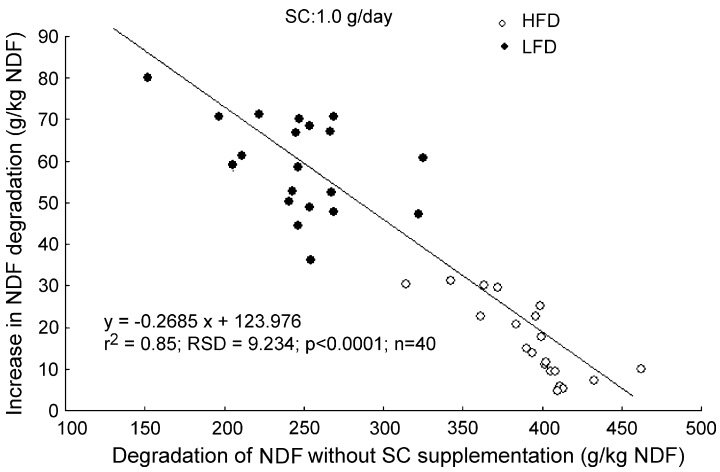

Fig. 3. Effects of supplementation with 1 g *Saccharomyces cerevisiae* per day (SC1.0) on fibre (NDF) degradation of maize silages after 36 h of incubation in the rumen of cows: HFD, high fibre degradation group and LFD, low fibre degradation group.

can increase fibre degradation of the diet, this demonstrates that both initial digestibility of silages, and level of yeast inclusion in the diet, will affect the response.

5. Conclusions

Feeding 0.3 or 1.0 g/day of SC (1×10^{10} CFU) in a diet of maize silage, concentrate and hay (48:42:10, DM basis) to non-lactating cows close to energy maintenance alleviated pH depression that typically occurs after feeding, reduced rumen lactate concentration and acetate:propionate ratio, but had no effect on the diurnal pattern of ammonia N concentration in ruminal fluid. The highest level of SC supplementation increased VFA concentration over the control diet by 17.9%, further reduced acetate:propionate ratio and increased fibrolytic activity of rumen bacteria as assessed by ruminal *in sacco* NDF degradation of 40 samples of maize silage differing in degradability. This effect was ($r^2 = 0.85$) inversely correlated with initial digestibility of silages. Results show that this *Saccharomyces cerevisiae* has the potential to reduce the risk of rumen acidosis in commercial cattle production and, if used at the appropriate amount, may increase metabolizable energy available from low quality maize silages, and the glucogenic potential of the diet, both of which would increase the efficiency of cattle production.

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