academicJournals

Vol. 10(53), pp. 4888-4895, 31 December, 2015
DOI: 10.5897/AJAR2015.10520
Article Number: 602880856581
ISSN 1991-637X

http://www.academicjournals.org/AJAR

African Journal of Agricultural Research

Full Length Research Paper

Effect of inactivated yeast on rumen dry matter degradation and fermentation of low concentrate feed

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Received 13 October, 2015; Accepted 20 November, 2015

Life yeast is known to improve rumen fermentation characteristics and performance, especially with high concentrate diet, while reports on inactivated yeast are ambiguous. Therefore, the present study focused on short-term effects of inactivated yeast (Levabon®) with an inclusion rate of 0 or 50 g per head per day on *in situ* rumen dry matter degradability (ISDMD) of a total mixed ration (66.6% grass silage, 14.5% maize silage, 6.3% soybean meal, 6.3% wheat and 6.3% barley) and its individual feedstuffs at low level of feed intake (7 kg/day) and at low concentrate diet. Eight non lactating rumen fistulated dairy cattle were used in a 4 × 2 Latin square designs in two identical periods (each period extended for 23 days) to measure *in situ* rumen degradation characteristics. Samples from the ruminal fluid were collected for further investigation (ruminal pH value, volatile fatty acids (VFAs), and ammonia nitrogen as well as acetate propionate ratio). The obtained results of this experiment indicated that Levabon® had no effect on rumen dry matter degradability of the total mixed ration and its individual feedstuffs. Levabon® had no effect on rumen dry matter degradability parameters and effective rumen dry matter degradability of the different feedstuffs. Moreover, rumen physiological parameters (rumen pH values, volatile fatty acid (VFAs) and ammonia nitrogen, as well as acetate propionate ratio), was not affected due to the addition of Levabon®. In conclusion, under the present feeding conditions (low level of feed intake and high proportion of structured roughage), Levabon® had no effect on rumen dry matter degradability or rumen physiological parameter due to absence of dietary factors challenging rumen fermentation.

Key words: Levabon®, rumen manipulation, rumen fermentation, *in situ* method, ammonia, volatile fatty acids.

INTRODUCTION

In the last few decades, feed additives, such as antibiotics, ionophores, methane inhibitors and defaunating agents have been used to manipulate and improve rumen fermentation and whole ruminant performance. Due to the ban in the use of antibiotic, much effort has been devoted towards developing alternatives to

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antibiotics (Anadón, 2006). Yeast as a direct feed microbe for cattle received a special attention (Chaucheyras-Durand et al., 2008). Nowadays, yeast products, either life or inactivated, are widely used as feed additives in ruminant animals, especially high producing dairy cows to improve rumen fermentation and performance. Yeast cells are known to be a rich source of vitamins, enzymes and some unidentified cofactors that seem to be helpful in increasing microbial activity in the rumen (Dawson et al., 1990). Previous studies tried to explain the different mechanisms by which yeast might act to improve ruminant production. Firstly, through improvement of rumen fermentation life, yeast cell was suggested to act as oxygen scavenger (Miller-Webster et al., 2002; Chaucheyras-Durand et al., 2008); stimulate lactic acid utilization (Callaway and Martin, 1997), to stabilize rumen pH (Newbold et al., 1996), increase production of volatile fatty acids and to alter its proportions (Williams et al., 1991).

Secondly, through improvement of rumen microbial population and activity there is increase in cellulolytic, pectinolytic and total bacteria (Dawson et al., 1990), ruminal protozoa population (Ayala et al., 1992), and total microbial protein production (Beauchemin et al., 2003). While life yeast culture is known to improve rumen fermentation characteristics and performance (Chevaux and Fabre, 2007), especially with high concentrate diet (Michalet-Doreau et al., 1997), reports on inactivated yeast are ambiguous. Previous studies reported that addition of inactivated yeast to ruminant diet had significant (Oeztuerk et al., 2005 and 2009) as well as non-significant (Opsi et al., 2012) effects on rumen fermentation and performance. Oeztuerk et al. (2005) attributed those differences to the differences in yeast strain, commercial products or dietary compositions. Therefore, the present study focused on short-term effects of inactivated yeast *in situ* rumen dry matter degradability (ISDMD) of a total mixed ration (TMR) and its individual feedstuffs at low level of feed intake and low concentrate diet. Rumen physiological parameters (rumen pH values, volatile fatty acid, and ammonia nitrogen as well as acetate propionate ratio) was also investigated to characterize possible mechanisms by which inactivated yeast might affected rumen fermentation.

MATERIALS AND METHODS

The present work was conducted at Chair of Animal Nutrition, Center of Life and Food Sciences, Weihenstephan, Technische Universität München, Germany. Dry matter degradability (DMD) of TMR and the individual components (grass silage, maize silage, soybean meal, wheat and barley) with or without inactivated yeast (Levabon®, Erber AG, Tulln, Austria) supplementation (0 or 50 g per head per day as company recommendation) was studied using the nylon bag technique (Ørskov and McDonald, 1979). In contrast to common *in situ* studies (protein degradability of distinct feed components added to the ration), the study also focused on the impact of the inactivated yeast additive on rumen fermentation kinetics of the entire ration. Another aspect was preparation of feed samples. Usually, the feed samples are dried and ground but this might modulate fermentation kinetics inside the nylon bag compared to the situation outside the bag. Therefore, the feed sample preparation was done with fresh materials.

Animals and diets

Eight non-lactating Friesian dairy cows (live body weight approximately 650 kg) were used to measure *in situ* rumen degradation characteristics. The cows were provided with rumen cannula (Bar Diamond Inc., Parma. Idaho, USA with 10 cm internal width). During the *in situ* experimental period, the cows were individually penned in clean and full automatic aerated stall (temperature 20°C). Daily dry matter intake was about 7.0 kg and the cows were given the ration in two equal portions at 07.00 am and 04.00 pm. Each portion on DM basis consisted of 2.33 kg from grass silage, 0.5 kg maize silage, 0.22 kg soybean meal, 0.22 kg wheat, 0.22 kg barley and 25 g mineral and vitamin mixtures. The yeast product purchased from BIOMIN Erber AG, Tulln, Austria (Levabon®) was added individually at every meal to the yeast treated cows in proportion of 7 g/kg DM (25 g per meal) as top dressing and thoroughly mixed to the other feed ingredients. Clean fresh water and salts blocks were offered for free choice. Ration was given for 10 days before the start of the experiment for adaptation (pre-experimental phase) and extended throughout the experimental period (13 days, experimental phase).

Fistulated cows were used in a 8 × 2 Latin square with factorial arrangement of treatment in two periods. During the first period, 4 cows received control treatment (no Levabon®) while the other animals were exposed to dietary Levabon® addition. In the second period, the treatment was reversed, thus providing each cow to serve as its own control. The care, maintenance, handling and surgical techniques of the animals were carried out according to the guidelines of the German laws for animal care.

Rumen incubation

The used bags (10 × 20 cm) had a pore size of 53 µm (R1020, Dohod Technology, Fairport. NY, USA). Four grams of DM of each (13.5 g fresh grass silage, 11.5 g fresh maize silage, 4.6 g from each; soybean meal, wheat or barley and 11.5 g from TMR) were weighed to the nearest 3 decimal points. The weighed materials were placed into previously labelled, dried (at 60°C for 48 h) and weighed bags, which were incubated in the rumen of the fistulated cows. For TMR bags the individual components were weighed and placed inside the bags in the same proportion as present in the ration and thoroughly mixed. In order to guarantee homogeneous presence of Levabon® in all tested material, Levabon® was added also to the material inside the bags in the same proportion as it was present in the respective TMR and its components. Nylon bags of the control treatment received no Levabon®.

Twenty four bags were prepared for each cow at each incubation point (4 bags from each; grass silage, maize silage, soybean meal, wheat, barley and TMR). Additionally, eighteen “0-hour” nylon bags were prepared (3 bags for each treatment) to serve as control at each incubation time. Dry mater content of feed material used was determined for each incubation time. The test-bags were incubated in the rumen of the eight cows just before morning feeding at 07.00 am for 1, 2, 3, 4, 5, 6, 9, 12, 24 and 48 h. Bags were removed from the rumen (all in – all out system) and were immediately put in ice water to stop microbial activity. Then the bags were put together with the corresponding 0-h bags into the washing tank with about

Table 1. Chemical composition of the different feedstuffs.

Feedstuff	DM (%)	(%) DM								
		OM	CP	EE	NfE	CF	Hemicell	Cellulose	Lignin	Ash
TMR	-	90.2	17.50	3.95	42.5	25.4	16.6	20.8	2.65	8.73
Grass silage	29.6	87.3	13.2	2.87	36.9	31.5	20.9	26.7	2.86	11.5
Maize silage	38.7	95.9	5.82	3.48	59.6	21.8	17.5	21.2	1.83	2.88
Soybean	88.8	94.2	48.7	1.70	34.4	7.87	4.45	6.14	0.43	6.87
wheat	88.3	94.6	14.7	1.30	36.3	3.33	8.49	12.2	7.46	1.90
Barley	89.2	97.9	9.91	1.92	76.3	5.30	6.48	2.49	0.87	2.18

40 L cold water and washed for about 5 min and then washed in a washing machine (QUELLE WVA BASIC 74) for 19 min. Afterwards the bags were freeze dried and weighed again to determine the *in situ* dry matter degradability.

Ruminal fluid samples (about 200 ml) were collected from each animal at the short term incubation periods (1, 2, 3. 4, 5, 6 and 9 h) and at the onset of incubation and at removal of nylon bags. Samples were divided into two portions; one portion was used directly to measure rumen pH and then was centrifuged, frozen and later used to measuring ammonia nitrogen (NH_3N). The second part was centrifuged and 10 ml of the supernatant was preserved and frozen to determine VFAs later on.

Chemical analysis

Samples of the feedstuffs (with and without Levabon® addition) were collected during the course of the study and then, pooled, dried, ground and submitted for chemical analysis. Chemical composition (Table 1) which included dry matter (DM), crude ash (CA), crude protein (CP), ether extract (EE) crude fiber (CF), fiber fractions in form of organic neutral detergent fiber (NDF), organic acid detergent fiber (ADF), acid detergent lignin (ADL) was carried out by weender analysis according to the standard procedures of VDLUFA (2004). Rumen fluid pH values were immediately measured using a pH-meter (Schott, CG 842). Analysis of ammonia nitrogen was conducted by a modified method of Conway (VDLUFA, 1976). Determination of rumen juice volatile fatty acids (acetate, propionate, butyrate and valeric) was done according to the method of Geissler et al. (1976).

Rumen degradation kinetics

Rumen dry matter degradation data were fitted to the exponential equation of Ørskov and McDonald (1979).

$$p = a + b(1 - e^{-c(t-t_0)}) \text{ for } t = t_0 \quad (1)$$

Where, P = DM degradation (%) at time t, a = rapidly soluble fraction (%), b = insoluble but ruminally degradable fraction (slowly degradable)(%), c = constant rate of degradation of b (%/h), t_0 = lag time (h), defined as the time from beginning of incubation until beginning of degradation (delay time). Effective rumen DM degradability (EDMD) were calculated following the equation of McDonald (1981).

$$P = a + \left[(b \times c)/(c + k)\right] \times e^{-k \times t_0} \quad (2)$$

Where a, b, c and t_0 are the same as in (1), k (%h^{-1}) is the estimate rate of passage of the digesta from the rumen per hours. The effective rumen dry matter degradability was calculated assuming a passage rate of 6% h^{-1}.

Statistical analysis

Average DM losses from bags within cows, treatment and incubation intervals, as well as corresponding rumen fluid pH values were subjected to analysis of variance with GLM procedures of SAS (SAS Institute Inc., Vers. 9.2):

Yij = μ + treatmenti + cowj + eij

Where, Yij = observation value of the dependant variable, μ = overall mean, treatmenti = fixed effect of Levabon® treatment (no vs. yes), cowj = fixed effect of cow (8 animals), eij = residual error. Differences between treatment (Levabon® addition: no vs. yes) were assessed for statistical significance by F-Test (treatment vs. eij) ($p < 0.05$).

RESULTS

In situ rumen dry matter degradability of the different feedstuffs

In situ rumen dry matter degradability of the different feed ingredients with or without Levabon® addition is presented in Table 2. The obtained data indicated that addition of Levabon® did not affect *in situ* rumen dry matter degradability of TMR and its individual components. Indeed, some isolated spots with statistically significant differences were detected but without quantitative relevance. A considerable proportion of variation in short term rumen fermentation kinetics was caused by individual differences among cows (see SD in Table 2). Repeatability of measurements within cows, however, was rather high, thus producing almost identical means for dry matter degradability between the two treatments with comparably low standard deviation of individual values (RMSE) and means (SEM),respectively.

Table 2. *In-situ* rumen dry matter degradability (ISDMD) of the different feedstuffs.

Feedstuff		Incubation time (h)										
		0	1	2	3	4	5	6	9	12	24	48
	-	38.1	41.1	41.2	42.9	44.2	46.1	48.4	53.1	57.5	75.1	81
	+	37.6	40.8	41.2	43.2	43.6	45.6	48.8	53.7	58.5	73.5	80.8
TMR	SD	-	0.70	0.69	1.39	1.22	2.35	2.82	3.48	5.10	1.89	1.05
	RMSE	-	0.83	0.69	1.75	3.06	2.05	2.46	3.85	6.44	3.54	1.10
	SEM	-	0.31	0.26	0.66	1.16	0.78	0.93	1.46	2.43	1.34	0.42
	-	35.3	35.1[b]	35.2	37.0	37.5	39.4	41.2	46	52.1	69.4	77.5
	+	35.4	36.6[a]	36.1	37.5	37.5	39.3	42.8	47.3	51.4	68.9	77.1
Grass silage	SD	-	0.62	0.60	0.58	0.76	1.32	1.90	3.28	4.50	2.52	0.71
	RMSE	-	0.78	0.88	1.01	2.29	2.28	2.34	2.09	4.67	4.25	1.97
	SEM	-	0.29	0.33	0.38	0.87	0.86	0.89	0.79	1.77	1.60	0.74
	-	51.4	52.9[a]	51.7	52.4[a]	49.7	52.1	53.1	55.1	58.7	71.3	76.9
	+	50.8	50.8[b]	50.3	50.2[b]	49.6	50.3	53.4	55.0	58.1	68.8	77.1
Maize silage	SD	-	1.20	1.15	1.30	1.57	2.21	3.11	2.86	4.69	1.51	0.64
	RMSE	-	1.30	2.03	1.52	4.50	3.18	1.27	2.42	2.95	2.48	2.05
	SEM	-	0.49	0.77	0.57	1.70	1.20	0.48	0.91	1.12	0.94	0.77
	-	27.4	30.0	30.5	32.9	35.1	38.6	40.9	54.2	64.8	93.8	97.9
	+	28.1	30.4	31.3	33.0	36.1	37.5	41.9	54.8	68.7	92.2	97.7
Soybean	SD	-	0.90	0.64	0.99	0.96	3.34	3.87	6.02	7.64	1.65	0.18
	RMSE	-	2.35	2.28	2.94	3.10	2.43	3.60	6.73	8.01	2.39	0.36
	SEM	-	0.88	0.86	1.11	1.19	0.92	1.36	2.54	3.03	0.90	0.14
	-	44.6	64.2	69.5	72.0	74.0	74.2	75.5	81.8	87.1	93.0	93.4[b]
	+	47.0	64.0	69.5	72.2	72.3	74.2	81.0	80.0	85.5	93.5	93.9[a]
Wheat	SD	-	2.29	2.40	2.94	3.86	5.66	7.11	5.76	5.11	0.36	0.28
	RMSE	-	6.08	3.34	6.25	5.08	7.42	5.80	7.93	2.76	0.78	0.23
	SEM	-	2.30	1.26	2.36	1.92	2.80	2.19	3.00	1.04	0.29	0.09
	-	22.6	42.6	48.3	59.2	59.4	62.7	67.7	74.5	80.8	90.0[b]	91.1[b]
	+	22.7	41.2	48.0	57.2	60.2	63.8	70.1	76.2	80.6	91.5[a]	92.9[a]
Barley	SD	-	3.58	4.47	5.00	3.79	6.72	3.09	5.74	6.60	0.46	0.38
	RMSE	-	6.98	7.45	7.60	10.75	6.15	5.30	8.92	5.10	0.90	0.53
	SEM	-	2.64	2.82	2.87	4.06	2.33	2.01	3.37	1.93	0.34	0.20

"-"control treatment, "+" Levabon®; SD: standard deviation between cows; Means having different letter within the same column at each feedstuff are statistically different. RMSE: standard error; SEM: standard error of means; "0h" samples were analysed before incubation (no relevance of standard deviation).

In situ rumen dry matter degradation kinetics of the different feedstuffs

Results showed that addition of Levabon® had no significant difference either on the parameters of rumen dry matter degradability or the effective rumen dry matter degradability with 6% rate of passage (Table 3).

Rumen fermentation characteristics

Rumen physiological parameters (rumen fluid pH value, rumen ammonia nitrogen, rumen total volatile fatty acids and acetate propionate ratio) are illustrated in Table 4. The obtained results indicate that addition of Levabon® had no significant effect on rumen physiological parameters. Individual cows showed same patterns of pH values but at remarkably different levels. These levels remained fairly constant between treatments. Consequently, the standard deviation between cows was comparably high, while the average pH pattern of the two treatments showed an almost identical shape. Rumen total volatile fatty acids showed the same patterns of pH values and reflected it. Therefore, the pattern of rumen

Table 3. Rumen degradability parameters and effective rumen dry matter degradability of the different feedstuffs with (+) or without (-) Levabon® addition (bold values = estimated parameters, values below = standard deviation).

Feedstuff	d	a	b	c	t_0	Passage rate (k, %/h)
	(%)	(%)	(%)	(%h^{-1})	(h)	6%
TMR -	12.5	39.8	47.6	6.14	2.22	59.3
	6.14	076	6.59	2.90	1.66	2.08
TMR +	11.9	38.5	49.6	5.73	1.33	59.0
	7.61	1.21	8.33	2.56	1.42	2.41
Grass silage -	15.7	35.7	48.6	6.56	4.03	53.8
	9.74	0.61	9.95	2.81	1.39	1.77
Grass silage +	16.4	36.3	47.3	5.90	3.76	53.9
	7.11	0.53	7.09	2.44	1.08	1.83
Maize silage -	18.1	51.4	30.4	8.28	6.03	61.5
	8.79	1.44	8.45	6,87	2.83	1.17
Maize silage +	17.6	50.1	32.3	5.71	5.64	60.4
	8.13	0.96	8.25	2.33	3.00	2.10
Soybean meal -	0.22	30.4	69.4	9.76	4.08	63.7
	0.41	1.98	2.02	2.29	1.22	3.25
Soybean meal +	0.29	30.8	69.0	9.79	3.81	64.3
	0.58	0.83	1.14	2.60	0.66	2.98
Wheat -	7.75	50.4	41.6	23.0	0.00	82.2
	1.98	2.36	2.37	12.9	0.00	2.62
Wheat +	6.59	52.5	40.9	20.0	0.00	82.4
	1.72	3.33	2.84	11.2	0.00	3.06
Barley -	10.6	26.8	62.6	22.0	0.00	74.0
	3.46	2.19	4.55	12.9	0.00	2.78
Barley +	8.61	27.0	64.4	18.6	0.00	75.2
	1.38	1.81	3.04	4.85	0.00	2.72

d = Non degradable fraction (%), a = rapidly soluble fraction (%), b = insoluble but ruminally degradable (slowly degradable fraction) (%), c = constant rate of degradation of b (%/h), t_0 = lag time (h), defined as the time from beginning of incubation until beginning of degradation (delay time).

total volatile fatty acids was considered as such a mirror for the pattern of rumen pH value. Rumen total volatile fatty acids reached the peak after 2 h from feeding and this was reflected by lowest pH value at this point.

DISCUSSION

One of the characteristics of the experimental design of the present study was the use of each individual cow as its own control. This was very important for maximizing precision of measurements since individual levels of short term fermentation kinetics and rumen fermentation characteristics revealed to vary considerably between cows but to be rather persistent over time and treatment within each cow. Considering the fact that the time patterns of dry matter degradability and rumen fluid pH of the two treatments were almost identical, it may be

Table 4. Rumen fluid pH value, ammonia nitrogen and volatile fatty acids of the different cattle with (+) or without (-) Levabon® addition.

Parameter		Time							
		7:00	**8:00**	**9:00**	**10:00**	**11:00**	**12:00**	**13:00**	**16:00**
Rumen pH -	**-**	6.94	6.36	6.18	6.39	6.50	6.52	6.48	6.80
	+	6.88	6.33	6.22	6.32	6.47	6.42	6.50	6.81
	SD	0.11	0.15	0.29	0.24	0.22	0.20	0.31	0.16
	SEM	0.06	0.14	0.11	0.12	0.09	0.09	0.08	0.10
NH_3N (mg/L)	**-**	52.0	392.9	236.8	380.6	320.0	107.9	85.6	29.8
	+	53.0	407.9	231.8	401.9	247.5	101.2	93.5	33.3
	SD	0.11	282.9	52.3	256.3	213.2	38.9	22.6	15.9
	SEM	0.08	0.93	0.88	0.89	0.58	0.78	0.57	0.72
Total VFA (mg/ml)	**-**	6.91	8.96	8.7	9.56	8.50	7.99	6.64	6.56
	+	6.44	9.28	7.2	9.45	8.13	7.27	6.75	6.75
	SD	2.22	1.54	1.52	2.51	2.23	2.76	2.06	1.89
	SEM	0.73	0.73	0.15	0.94	0.79	0.67	0.93	0.87
Ace/Pro ratio	**-**	3.75	2.31	2.37	2.56	2.76	2.93	3.2	3.54
	+	3.84	2.28	2.51	2.56	2.88	2.98	3.2	3.58
	SD	0.37	0.09	0.34	0.18	0.19	0.23	0.13	0.17
	SEM	0.66	0.6	0.51	0.98	0.32	0.71	0.93	0.75
Lactic acid	**-**	0.008	0.584	0.137	0.014	0.007	0.008	0.012	0.006
	+	0.011	0.703	0.067	0.006	0.01	0.006	0.013	0.005
	SD	0.007	0.351	0.194	0.011	0.01	0.003	0.011	0.004
	SEM	0.56	0.584	0.556	0.28	0.556	0.293	0.856	0.655

concluded that the absence of a significant effect of inactivated yeast (Levabon®) addition on the aforementioned parameter was measured with rather high precision. Individual patterns of DMD of TMR and its individual components were typically as shown widely in literature (maize silage as an example, the comparably high content of quickly soluble material and the slow onset of degradation of its insoluble material) reflect this concept. These results are in agreement with Opsi et al. (2012) who studied the effect of adding live and autoclaved yeast on the *in vitro* dry matter degradability of the total mixed ration and concluded that both of the yeast products did not affect the *in vitro* dry matter degradability.

The results of the current work indicated that adding Levabon® had no effect on rumen pH values. Previous studies (Piva et al., 1993; Enjalbert et al., 1999; Lynch and Martin, 2002; Erasmus et al., 2005) are in agreement with the current work and has found that adding inactivated yeast culture had no effect on pH values when TMR with variable forage to concentrate ratios (ranging from 40:60 to 67:33%) were fermented. Moreover, ruminal NH_3-N concentration was not affected by adding Levabon®, which is consistent with previous studies (Lila et al., 2004; Erasmus et al., 2005; Guedes et al., 2008) using different substrates in their experiments. However, the current results disagreed with those reported by Lynch and Martin (2002), Oeztuerk et al. (2005) and Oeztuerk (2009), who found a decline in pH and increase in ammonia concentration due to long-term effects of adding autoclaved and live yeast to Rusitec fermenters, although the effects were more pronounced when live yeast culture was used. In all cases, the pH values remained within the physiological range of a healthy rumen.

The current work revealed that Levabon® addition had no effect on VFAs concentration and its molar proportion (acetate propionate ratio). These results are consistent with the previous aforementioned results of constant rumen pH due to addition of Levabon® as it is an indicator for rumen ammonia nitrogen and VFAs production. These results are compatible and in agreement with the results of Opsi et al. (2012) in the addition of both live and inactivated yeast culture to high

concentrate total mixed ration (60% concentrate to 40% roughage) using the *in vitro* method. It is well known that the production and the changing in VFAs concentration and its molar proportion is attributed to the changing of bacterial population. Acetate as the main volatile fatty acid is mainly formed due to structural carbohydrate fermentation by cellulolytic bacteria, whereas a relatively greater production of propionate is formed due to the fermentation of non-structural carbohydrate by amylolytic bacteria. Reports from the previous studies on microbial population modification that occur within the rumen in response to live yeast addition to the diet were found to increase (Newbold et al., 1996) the numbers of total viable bacteria, and increased (Wiedmeier et al., 1987; El Hassan et al., 1996) the counts of cellulolytic bacteria, with no effects (Kumar et al., 1994) on amylolytic bacteria. These reported trends towards an increased ratio of cellulolytic to amylolytic bacteria could therefore lead to a change in VFAs production and an increased acetate propionate ratio.

Conclusion

Under the present feeding conditions (low level of feed intake, low level of concentrate and high proportion of structured roughage), addition of inactivated yeast to ruminant diet had no effect, neither on *in situ* rumen degradation characteristics of dry matter or rumen fermentation characteristics.

Conflict of Interests

The authors have not declared any conflict of interests.

ACKNOWLEDGEMENT

The authors gratefully acknowledge and appreciate BIOMIN Erber AG, Tulln, Austria, for the financial support of this study.

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