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Animal Feed Science and Technology

journal homepage: www.elsevier.com/locate/anifeedsci



Short communication

Effect of forage source and a supplementary methionine hydroxyl analogue on rumen fermentation parameters in lactating dairy cows offered a low crude protein diet^{\ddagger}





S.J. Whelan^a, F.J. Mulligan^{b,*}, J.J. Callan^a, B. Flynn^a, K.M. Pierce^a

^a School of Agriculture and Food Science, University College Dublin, Lyons Research Farm, Newcastle, Dublin, Ireland
^b School of Veterinary Medicine, Veterinary Science Centre, University College Dublin, Belfield, Dublin 4, Ireland

ARTICLE INFO

Article history: Received 10 October 2011 Received in revised form 12 April 2013 Accepted 22 April 2013

Keywords: Forage source Methionine Rumen fermentation

ABSTRACT

The objective of this experiment was to evaluate the effects of forage source (grass silage (GS) or maize silage (MS)) and a supplementary methionine hydroxyl analogue (HMBi) on rumen fermentation parameters in dairy cows offered a low-crude protein (CP) diet. Four lactating dairy cows permanently fitted with rumen cannulae were assigned to 1 of 4 diets in a 2×2 factorial, Latin square design. Diets were isoenergetic and isonitrogenous and were offered once daily as a total mixed ration. Rumen samples were taken every 2 h over 48 h for analysis of pH, volatile fatty acids (VFA), total lactic acid and NH₃-N. Mean pH (6.21 ± 0.03) and nadir pH (5.64 ± 0.12) were not affected by dietary treatment. Animals offered GS based diets had higher VFA ($113.69 \times 107.55 \pm 1.53 \text{ mmol/L}$) and NH₃-N ($62.82 \times 55.54 \pm 2.38 \text{ mg/L}$) concentrations vs. those offered MS diets. No effect of supplementary HMBi on rumen pH, VFA or NH₃-N was detected in this study.

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1. Introduction

Dietary strategies to reduce N losses from dairy production have become more prominent due to environmental concerns. Strategies such as reducing dietary CP, alternative forage sources to grass silage (GS) (Burke et al., 2007) and offering supplementary amino acid (AA) analogues (Broderick et al., 2009) have been evaluated as a means of reducing N loss. However, there are few studies reporting the effects of these strategies on rumen physiology, despite this organ being essential for the efficient capture of nutrients and maintenance of animal health. For example, reducing CP by replacing protein with fermentable energy will reduce rumen NH₃-N and increase volatile fatty acid (VFA) concentrations thus reducing rumen pH (Briggs et al., 1957). In Northern Europe, GS and maize silage (MS) are the most common forages offered to dairy cows during the housing period (Rath and Peel, 2005). Grass silage contains slowly degradable energy, rapidly degradable N (McDonald et al., 2002) and is frequently supplemented with barley based concentrates. Such feeding situations may result in elevated NH₃-N and lactic acid concentrations contributing to poor N capture from the diet and depressions in rumen pH. By contrast, MS contains starch which is less extensively degraded than rolled barley (Owens et al., 2009) and is lower in CP than GS, potentially reducing both NH₃-N and lactic acid concentrations in the rumen. Feeding low-CP diets may result in an AA

Abbreviations: dNDF, digestible neutral detergent fibre; GS, grass silage; HMBi, isopropyl ester of a methionine hydroxyl analogue; MS, maize silage; T, time; TMR, total mixed ration; VFA, volatile fatty acids.

^{*} Funding for this research was provided under the National Development Plan, through the Research Stimulus Fund, administered by the Department of Agriculture, Fisheries & Food, Ireland#RSF 07536.

^{*} Corresponding author. Tel.: +353 17166251; fax: +353 16288421. *E-mail address*: finbar.mulligan@ucd.ie (F.J. Mulligan).

^{0377-8401/\$ -} see front matter © 2013 Elsevier B.V. All rights reserved. http://dx.doi.org/10.1016/j.anifeedsci.2013.04.017

Table 1

Chemical composition of dietary treatments offered.

	Forage		Dietary treatments			
	Grass silage	Maize silage	GS- ^a	GS+ ^a	MS- ^a	MS+ ^a
Chemical composition (g/k	g DM unless stated)					
DM (g/kg)	222	287	363	365	400	410
ME (MJ/kg DM)	10.3	11.3	11.31	11.30	11.54	11.48
DMD ^b	670	695	744	725	730	722
Crude Protein	108	89	133	132	133	134
Lysine	3.4	2.5	5.4	5.4	5.3	5.3
Methionine	1.2	1.1	1.5	2.0	1.5	2.0
Protein fractions						
Fraction A (g/kg CP)	690	510	451	451	399	399
Fraction B (g/kg CP)	200	298	444	444	457	457
Fraction C (g/kg CP)	7	10	29	29	28	28
Starch	-	263	146	147	178	177
NDF	650	490	500	514	591	583
NDF _{forage} ^c	650	490	288	288	315	315
ADF	240	273	216	205	223	229

^a GS- = grass silage based total mixed ration (TMR), GS+ = grass silage based TMR with supplementary 2-hydroxy-4-methylthio butanoic acid isopropyl ester (HMBi), MS- = maize silage based TMR, MS+ = maize silage based TMR with supplementary HMBi.

^b DMD = digestibility of DM.

^c NDF_{forage} = contribution of the forage component of the diet to NDF.

deficiency, of which methionine is said to be first limiting in dairy cows (NRC, 2001). Whilst crystalline forms of methionine are rapidly degraded in the rumen; offering a methionine hydroxyl analogue (isopropyl ester of 2-hydroxy-4-methylthio butanoic acid, HMBi) infers a degree of rumen protection, improving methionine supply to the animal (Graulet et al., 2005). Previously, Noftsger et al. (2005) demonstrated improvements in rumen fibre digestion where dl-methionine or HMBi were offered to dairy cows. However, an effect of HMBi on rumen pH, VFA or NH₃-N has yet to be elucidated. Thus, the objective of this experiment was to evaluate the effects of offering either GS or MS based diets with or without supplementary HMBi on rumen pH, VFA and NH₃-N concentrations in lactating dairy cows offered low-CP diets.

2. Materials and methods

All procedures involving the use of animals were conducted under experimental licence from the Department of Health and Children, Ireland (S.I.613, 2005). Four multiparous, Holstein Friesian dairy cows (125 ± 4 d in milk and 23.5 ± 2.3 kg/d pre-experimental milk yield), permanently fitted with rumen cannulae (Bar Diamond Inc. Idaho, USA) were assigned to 1 of 4 dietary treatments in a 2 × 2 factorial, Latin square design. Each experimental period allowed 14 d for dietary acclimatisation followed by 7 d where animals were housed in individual metabolic stalls; 2 d for stall acclimatisation and 5 d for sampling. Animals were milked twice daily and milk yield was recorded (Weighall Milk Meters; Dairy Master, Kerry, IE). Diets were offered once daily at 08:00 as total mixed rations (TMR) with allowances made for refusals of 0.1. Mixing was achieved using an experimental diet feeder (Data Ranger, American Callan, NH, US). Grass silage consisted of predominantly *Lolium perenne* whilst MS consisted of *Zea mais cv*. Justina. The management of GS and MS used in this experiment has been reported in a companion study by Whelan et al. (2011). Diets contained either predominantly GS (0.8 of forage DM, GS+ and GS-) or MS (0.8 of forage DM, MS+ and MS-) and were offered with (1.2 g/kg DM; GS+ and MS+) or without (GS- and MS –) HMBi (Metasmart Dry, Adisseo, Commentry, France). Each TMR contained 440 and 340 g/kg DM concentrate for GS and MS based diets respectively. The chemical composition of each diet is presented in Table 1.

Feed offered, feed refusals and faecal output were weighed and sampled daily for 5 d. Samples of each TMR and faeces were dried at 55 °C in a forced air oven for 48 h and 96 h respectively. Milk yield was recorded daily and a representative sample was harvested during am and pm milking and mixed according to output. Rumen fluid samples were harvested every 2 h over 48 h from the ventral sack (*Saccus ventralis*) of the rumen using a collection tube (#RT, Bar Diamond Inc. Idaho, US) and a 60 mL disposable syringe. The pH was immediately measured (pH3100, WTW, Weilheim, DE.) and an 8 mL subsample was mixed with 2 mL trichloroacetic acid (500 g/L) prior to storage at -20 °C. Dried feed and faeces were prepared and analysed for NDF, ADF, starch, ash and N as described previously in Whelan et al. (2011), whilst dietary CP fractions were determined according to Licitra et al. (1996). Nutrient digestibility was determined by total faecal collections as described by Mulligan et al. (2002a) whilst milk samples were analysed for fat protein and lactose in a commercial laboratory (Progressive Genetics LTD, Dublin, IE) using infrared analysis (CombiFoss 5000; Foss Analytical A/S, Hilleroed, DK). Samples of rumen fluid were analysed for NH₃-N using the phenol-hypochlorite method of Weatherburn (1967) whilst VFA concentrations were determined using gas chromatography as described in Hart et al. (2009).

Data were checked for normal distribution and homogeneity of variance using histograms and statistical tests in Proc UNIVARIATE of SAS (2004). The portion of observations at which rumen pH was below 5.8 was calculated using Proc FREQ of SAS (2004). Analysis of data was carried out using the repeated measures function in Proc MIXED of SAS (2004) using the

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