BRIEF COMMUNICATION: Methane emissions by sheep offered high-sugar or conventional perennial ryegrass at two allowances

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Introduction

Perennial ryegrass (*Lolium perenne* L.) with a higher concentration of water-soluble carbohydrates (WSC) has been bred by plant breeders (Humphreys 1989). Ryegrass with a higher WSC can potentially alter rumen microbial fermentation, synchrony of energy and protein in the rumen, total energy supply to the animal and consequently productivity (Edwards et al. 2007). Methane originates mainly from rumen fermentation and is the main greenhouse-gas produced by New Zealand (NZ) agriculture (MfE 2013). The higher concentration of WSC in high sugar ryegrass (HSG) can change rumen fermentation patterns (Lee et al. 2003) and may consequently affect methane emissions. Methane emissions are largely driven by dry matter intake (DMI) (Hammond et al. 2013) and methane mitigation strategies are often more effective at a high DMI (Moe & Tyrrell 1979). However, a true methane mitigation strategy should be effective at any DMI level.

The objective of this study was to determine methane emissions from sheep fed freshly cut diploid HSG, conventional diploid ryegrass (CRG) and tetraploid ryegrass (TRG) offered at two levels of metabolisable energy for maintenance (MEm).

Materials and methods

An indoor trial was performed at AgResearch Grasslands (Palmerston North, NZ) from September 23 to October 11, 2013 with 48 Romney wether sheep of approximately one year of age (mean live weight = 33.3±2.07 kg). The sheep were grouped by weight and randomly allocated to one of six dietary treatments. Three perennial ryegrass cultivars, AberMagic (HSG), Alto (CRG) and Base (TRG) were fed at 1.1 or 1.8 × MEm (CSIRO 2007) to eight sheep per treatment. Grass at a vegetative stage was cut daily around noon and 65% of the total daily grass diet fed at 15:30 h and the remaining 35% stored refrigerated at 4°C until feeding the following morning around 8:30 h. Subsamples were collected daily in triplicate and dried at 105°C for 48 h to determine the dry matter (DM) content. During the methane measurement period, another daily subsample was taken, freeze-dried, ground through a 1 mm screen and scanned by near infrared reflectance spectroscopy (NIRS; feedTECH, AgResearch Ltd., Palmerston North, NZ) with calibration curves for ash, lipids, crude protein (CP), neutral detergent fibre (NDF) and WSC (Corson et al. 1999).

Methane emissions and dry matter intake (DMI) measurements were performed after acclimatisation of at least five days indoor housing and two days in individual metabolism crates before entering open circuit respiration chambers for two consecutive days (Pinares-Patiño et al. 2012). Four hours after morning feeding, on the day before the sheep entered the respiration chambers, a rumen sample was taken from each sheep via stomach tubing. The rumen samples were later analysed for volatile fatty acid (VFA) composition by gas chromatography as described by Sun et al. (2012). The ratio between VFAs that result in a net hydrogen production:net hydrogen reduction (h-VFAr) in the rumen was calculated as (acetate + butyrate)/(propionate + valerate) (Demeyer 1991).

Statistical analysis was performed for DMI, methane emissions and h-VFAr by two-way analysis of variance with fixed effects of ryegrass cultivar and feeding level and interaction between cultivar and feeding level using GenStat Version 13 (Payne et al. 2009). Sheep within dietary treatment was the experimental unit (n=8). The Tukey statement in GenStat was used for multi-treatment comparison and significance was declared at P <0.05.

Table 1 Nutritional composition (mean ± standard deviation; n=4) of high sugar diploid ryegrass (HSG), control diploid ryegrass (CRG), and tetraploid ryegrass (TRG) fed to sheep during the respiration chamber period.

	Ryegrass cultivar		
	HSG	CRG	TRG
Dry matter (g/kg)	166±33.4	167±30.5	130±13.9
Water-soluble carbohydrates (g/kg DM)	259±27.5	221±40.7	202±10.5
Crude protein (g/kg DM)	139±20.4	139±20.9	169±7.0
Neutral detergent Fibre (g/kg DM)	430±23.1	468±26.6	451±21.7
Ash (g/kg DM)	75±4.8	79±7.1	87±3.6
Lipid (g/kg DM)	23±0.4	20±2.2	23±2.4

Table 2 Dry matter intake, methane (CH_4) emissions and rumen fluid characteristics of sheep (n=8) fed one of three ryegrass cultivars (*cv.* HSG, high-sugar diploid ryegrass; CRG, control diploid ryegrass; TRG, tetraploid ryegrass) at one of two intake levels of maintenance metabolisable energy requirements (MEm). SED, standard error of the difference.

		Cultivar[1]				Cultivar		MEm		Cultivar×MEm	
	MEm	HSG	CRG	TRG	MEm mean	SED	P value	SED	P value	SED	P value
DM intake (kg/d)	1.1×	0.72	0.72	0.57	0.67	0.036	**0.006**	0.0291	**<0.001**	0.050	0.229
	1.8×	0.98	0.93	0.90	0.94						
	cv. mean	0.85^{b}	0.82^{b}	0.74^{a}							
CH_4 production (g/d)	1.1×	14.1^{xy}	15.9^{yz}	12.1^{x}	14.0	0.72	**0.044**	0.59	**<0.001**	1.02	**0.041**
	1.8×	17.2^{z}	17.4^{z}	17.4^{z}	17.4						
	cv. mean	15.6^{ab}	16.6^{b}	14.8^{a}							
CH_4 yield (g/kg DMI)	1.1×	19.7	22.3	21.4	21.1	0.66	**0.024**	0.54	**<0.001**	0.93	0.426
	1.8×	18.1	19.0	19.5	18.9						
	cv. mean	18.9^{a}	20.7^{b}	20.4^{ab}							
Volatile Fatty Acids (*mM*)	1.1×	82.4	75.5	69.3	75.7	2.55	**0.012**	2.08	**0.005**	3.60	0.120
	1.8×	84	80.3	81.4	81.9						
	cv. mean	83.2^{b}	77.9^{ab}	75.4^{a}							
(Acetate+Butyrate)/ (Propionate+Valerate) (i.e. h-VFAr)	1.1×	2.63	2.91	2.78	2.77	0.086	**<0.001**	0.070	**0.010**	0.122	0.156
	1.8×	2.31	2.65	2.78	2.58						
	cv. mean	2.47^{a}	2.78^{b}	2.78^{b}							

[ab]Mean ryegrass cultivar (*cv.*) within a single row with different superscripts are significantly different (P <0.05).
[xyz]Means within a block (interaction) with different superscripts are significantly different (P <0.05).

Results and discussion

The concentration of WSC was 38 and 57 g/kg DM higher in HSG compared with CRG and TRG, respectively (Table 1). The higher WSC in HSG was mainly at the expense of NDF compared with CRG (38 g/kg DM) and at the expense of CP compared with TRG (30 g/kg DM). Similar trends for HSG, CRG and TRG in terms of WSC, CP and NDF have previously been reported under NZ conditions (Cosgrove et al. 2007; Pacheco et al. 2009). Daily DMI was on average 0.67 kg DM/d at 1.1×MEm and 0.94 kg DM/d at 1.8×MEm (Table 2). At 1.1×MEm, DMI was lower for sheep fed TRG compared with sheep fed HSG or CRG. Feed allocation was based on fresh weight, and the lower DMI of TRG at 1.1×MEm reflects the lower DM content. Methane production was similar among cultivars (17.4 g methane/d) when sheep were fed at 1.8×MEm. At 1.1×MEm, sheep fed TRG produced less methane (P <0.05) compared with sheep fed CRG (HSG was intermediate), probably because intake was lower (P <0.05) for sheep fed TRG.

Methane yield (g/kg DMI) was on average 10% lower (P <0.001) for sheep fed at 1.8×MEm compared with sheep fed at 1.1×MEm. This trend of reduced

methane yield with increasing intake is commonly found in sheep (Sun et al. 2012; Hammond et al. 2013). Methane yield was 9% lower ($P < 0.05$) for sheep fed HSG compared with sheep fed CRG, with TRG fed sheep intermediate. This effect was observed at both intake levels. Methane yield was relatively low in this study compared to previous studies of the same institute with sheep fed fresh ryegrass which had an average methane yield of 24.3±2.1 g/kg DMI (Sun et al. 2012; Hammond et al. 2013). Methane is formed from hydrogen and carbon dioxide and therefore the h-VFAr gives an indication of methane production potential in the rumen (Demeyer 1991). The h-VFAr was lower for HSG ($P < 0.05$) compared with CRG and TRG, which is consistent with the lower methane yield.

Compared with the control diploid ryegrass, the high sugar ryegrass reduced methane yield by sheep at both feeding levels. This suggests a potential method for methane mitigation that could be readily adopted by NZ livestock farming. However, these results should be confirmed in additional trials in different seasons and under field conditions.

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