

The effect of different oils and diets on total gas production in an artificial rumen (Rusitec)*

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ABSTRACT

Four experiments were carried out in a Rusitec system. The effect of different diets (100% fresh lucerne, experiment I; 60% fresh lucerne + 40% maize, experiment II; 40% fresh lucerne + 60% maize, experiment III) and fat sources (linseed oil-LO, rapeseed oil-RO, fish oil-FO, 5%wt.wt⁻¹) on gas production and hydrogen balance was studied. In experiment IV the basic diet composition was as in experiment II but the diets were supplemented with 5%wt.wt⁻¹ of oil blends: LO+RO, LO+FO or LO+FO+RO. A significant ($P>0.05$) reduction in the total gas production was noticed only in the full forage diets supplemented with LO, RO and FO.

KEY WORDS: *in vitro*, lucerne, maize, oils, total gas, hydrogen

INTRODUCTION

In some developed countries special emphasis is placed on reduction of environmental degradation associated with animal production. Scientists have recently been searching for a new *in vitro* method that would reflect the feed fermentation profile in the rumen. Total gas production *in vitro* appears to be a potentially useful marker of feed component digestibility, e.g., starch fermentation (Chai et al., 2004). It is also possible to examine the effects of various fat types on *in vitro* gas production and diet digestibility (Getachew et al., 2001). The objective of the present study was to compare diets comprising three different fresh lucerne-

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to-maize ratios in diets supplemented with rapeseed, linseed or fish oils and their blends on total gas production in a Rusitec system.

MATERIAL AND METHODS

The rumen simulation technique was employed to test different types of diets (forage, forage:concentrate 60:40 or 40:60) and 5% wt.wt⁻¹ of oils (rapeseed RO, linseed LO, or fish oils FO) on total gas production and hydrogen balance. Details of the method were previously described by Jalč et al. (2006a). On days 8-13 gas samples were collected and the total volume was measured by a gas-meter. Production, utilization and recovery of metabolic hydrogen were calculated according to the stoichiometry of rumen fermentation as suggested by Demeyer (1991). The following equations were used:

a. production of metabolic H₂ = 2A + P + 4B + 2iV + 2V (mM day⁻¹)

b. utilization of metabolic H₂ = 2P + 2B + 4M + V (mM day⁻¹)

c. recovery of metabolic hydrogen = H₂ utilization/H₂ production × 100 (%),

where A, P, B, V, iV and M are the molar amounts of acetate, propionate, n-butyrate, valerate, isovalerate, and methane, respectively.

All data were analysed using one-way analysis of variance (ANOVA) and compared by the Tukey-Kramer multiple comparison test.

RESULTS AND DISCUSSION

The primary gaseous end product of fermentation is carbon dioxide and one-third of this is reduced to methane (Czerkawski, 1986). The gas mixture in the rumen consists largely of carbon dioxide (65%) and methane (26-27%). Some nitrogen (7%), traces of oxygen (0.5%) and some hydrogen (0.2%) may be also present. The amount of produced gas is proportional in volume to volatile fatty acid production, thereby serving as an indicator of volatile fatty acids produced by fermentation. In experiment I, total gas production was significantly (P<0.05) suppressed by oil supplementation to a forage diet (Table 1). In contrast, supplementation of oils and oil blends to high-forage (experiments II, IV) or high concentrate (experiment III) diets did not affect total gas production. Similar results were obtained by Getachew et al. (2001) when saturated fats (tallow or yellow grease) were tested. In the same study, however, potassium soaps of maize oil, tallow and yellow grease depressed gas production *in vitro*. The authors suggested that triglycerides have a much smaller effect on rumen fermentation parameters than the corresponding free fatty acids. On the other hand, oils rich in unsaturated fatty acids (e.g., maize oil) may stimulate elevated gas production in comparison with oils rich in saturated fatty acids (Getachew et al., 2001).

Although hydrogen is one of the major end products of fermentation in protozoa, fungi, and pure monocultures of some bacteria, it is not accumulated in the rumen, because it is immediately used by other bacteria present in the mixed microbial ecosystem. Metabolic hydrogen in the form of reduced protons (H) can

Table 1. Effect of different oils and their blends supplemented to diets on gas production in Rusitec (n-6)

Items	Added oil (5% wt · wt ⁻¹)				pooled ± SEM
	control ^a	LO ^b	RO ^c	FO ^d	
<i>100% fresh lucerne (experiment I)</i>					
Gas production, ml · day ⁻¹	3630.0 ^{b,c,d}	3341.0 ^{c,d}	3492.0 ^d	3468.0	56.4
H ₂ production, mM · day ⁻¹	80.52	74.91	83.49	75.75	2.1
H ₂ utilization, mM · day ⁻¹	26.36	30.81	28.87	31.32	1.4
H ₂ recovery,%	32.75 ^{b,c,d}	41.16 ^c	34.74	41.27	1.3
<i>60% fresh lucerne + 40% maize (experiment II)</i>					
Gas production, ml · day ⁻¹	3871.0	3698.0	3770.0	3701.0	48.2
H ₂ production, mM · day ⁻¹	88.32	82.48	87.64	81.76	2.3
H ₂ utilization, mM · day ⁻¹	29.31 ^b	36.22 ^c	29.88	32.16	1.5
H ₂ recovery,%	32.16 ^{b,d}	44.03 ^{c,d}	34.14 ^d	39.65	1.42
<i>40% fresh lucerne and 60% maize (experiment III)</i>					
Gas production, ml · day ⁻¹	3330.0	3597.0	3683.0	3416.0	36.8
H ₂ production, mM · day ⁻¹	77.65	77.75	84.69	74.09	2.2
H ₂ utilization, mM · day ⁻¹	26.23	32.10	29.49	26.86	1.1
H ₂ recovery,%	33.8	40.94 ^{a,d}	34.76 ^b	36.42	1.3
<i>40% fresh lucerne and 60% maize (experiment IV)</i>					
Gas production, ml · day ⁻¹	3754.0	3522.0	3741.0	3649.0	39.7
H ₂ production, mM · day ⁻¹	95.71 ^b	79.13	86.20	88.84	2.30
H ₂ utilization, mM · day ⁻¹	33.16	32.82	35.21	32.07	1.20
H ₂ recovery,%	34.59 ^{b,c}	41.62 ^d	40.89	36.09 ^c	1.60

LO - linseed oil; RO - rapeseed oil; FO - fish oil
 values in a row with different superscripts (a,b,c,d) differ at P<0.05

also be used during synthesis of volatile fatty acids or incorporated into microbial organic matter (Moss et al., 2000). The principle sources of hydrogen are bacteria and protozoa producing acetic acid (Hegarty and Gerdes, 1998). Hydrogen is a waste fermentation product and when accumulated, rumen digestion is suppressed (Wolin et al., 1997). In the present study, production of metabolic hydrogen was affected only in experiment IV after applying oil blends (LO+RO). With respect to hydrogen utilization during fermentation, it was significantly increased only in experiment II with LO (about 7 percentage units). In general, the recovery of metabolic hydrogen significantly increased with LO, RO, FO (experiment I), LO, FO (experiment II), LO (experiment III), and LO+RO, LO+FO (experiment IV).

Our previous study also showed a significant increase in hydrogen recovery when microbial oil was used (Jalč and Čertic, 2005). According to Demeyer et al. (1995), other hydrogen acceptors have to be considered in order to explain the lower hydrogen recovery, which also was observed when rapeseed and linseed oils were examined (Machmüller et al., 1998).

CONCLUSIONS

Total gas production was significantly reduced in a forage diet by LO, RO, and FO, while these oils and their blends (LO+RO, LO+FO, LO+FO+RO) added to diets having different forage-to-concentrate ratios did not affect total gas release. The production and utilization of metabolic hydrogen were not affected by oils or their blends, however, the recovery of metabolic hydrogen was decreased mainly with LO, FO, and oil blends (LO+RO, LO+FO).

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