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## Potential for functional replacement of methanogenic bacteria by acetogenic bacteria in the rumen environment

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Ruminal fermentation is influenced by interspecies H<sub>2</sub> transfer in which methanogens enhance energy production by fermentative microorganisms by converting  $H_2$  to  $CH_4$ . However, the energy contained in CH<sub>4</sub> represents a 5 to 15% loss of apparent digestible energy loss to the animal and contributes to global methane emissions. The objective of our research is to determine if alternative H<sub>2</sub>-utilizing bacteria, acetogens are capable to replacing the interspecies H<sub>2</sub> transfer function of methanogens when methanogens are inhibited. We have isolated several acetogenic isolates, A10 and <sup>3</sup>H, with low hydrogen thresholds (Boccazzi et al, 1993, Proc Conf Rumen Funct, 22-28). The purpose of this research was to determine if they could utilize H<sub>2</sub> in a mixed culture system.

Serum bottles (120 ml) containing 0.35 g ground alfalfa hay (1 mm) were inoculated with 10 ml of a solution consisting of 60% anaerobic dilution solution:40% fresh rumen contents from a cow consuming a 60% concentrate:40% corn silage diet. Bromoethane sulfonic acid (BES, 5 mM) was added to all serum bottles except the controls. Treatments were : control, control + BES,  $_{3}$ H + BES and A10 + BES,

where the acetogenic cultures were added to give a final concentration of 4 x  $10^8$  acetogen cells/ml under a CO<sub>2</sub> atmosphere. Triplicate serum bottles for each treatment and time period were incubated at 37°C and shaken at 200 rpm. Hydrogen and CH<sub>4</sub> concentration in the headspace was measured by gas chromatography using thermal conductivity.

No CH<sub>4</sub> accumulated in serum bottles where BES had been added which indicates that 5 mM BES completely inhibited methanogenesis for up to 74 h. Hydrogen concentrations at 12 h were lower for cultures containing added acetogens. High levels of H<sub>2</sub> in the control of 12 h may have been due to low numbers of methanogens in the inoculum. Concentrations of H<sub>2</sub> were lower for control bottles and bottles containing acetogens at 24 and 74 h. Cultures containing acetogenic isolate <sup>3</sup>H consistently had lower H<sub>2</sub> concentrations than cultures containing acetogenic isolate A10. These data show that acetogens can functionally replace the role of methanogens in interspecies H<sub>2</sub> transfer and that different levels of H<sub>2</sub> control may be obtained by using different acetogenic cultures.

Treatment		Η <sub>2</sub> (μΜ)		CH <sub>4</sub> (μM)	
	0h	12h	24h	74h	74h <sup>1</sup>
Control	10	166	16	0.5	258
Control + BES	8	171	112	23	0.8
3H + BES	8	48	12	2	1
A10 + BES	9	108	44	21	3

<sup>1</sup> Initial CH<sub>4</sub> concentrations were less than 2 µM.