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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_2 transfer in which methanogens enhance energy production by fermentative microorganisms by converting H_2 to CH_4 . However, the energy contained in CH_4 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_2 -utilizing bacteria, acetogens are capable to replacing the interspecies H_2 transfer function of methanogens when methanogens are inhibited. We have isolated several acetogenic isolates, A10 and 3H , 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_2 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, 3H + BES and A10 + BES,

where the acetogenic cultures were added to give a final concentration of 4×10^8 acetogen cells/ml under a CO_2 atmosphere. Triplicate serum bottles for each treatment and time period were incubated at $37^\circ C$ and shaken at 200 rpm. Hydrogen and CH_4 concentration in the headspace was measured by gas chromatography using thermal conductivity.

No CH_4 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_2 in the control of 12 h may have been due to low numbers of methanogens in the inoculum. Concentrations of H_2 were lower for control bottles and bottles containing acetogens at 24 and 74 h. Cultures containing acetogenic isolate 3H consistently had lower H_2 concentrations than cultures containing acetogenic isolate A10. These data show that acetogens can functionally replace the role of methanogens in interspecies H_2 transfer and that different levels of H_2 control may be obtained by using different acetogenic cultures.

Treatment	H_2 (μM)				CH_4 (μM)
	0h	12h	24h	74h	74h ¹
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

¹ Initial CH_4 concentrations were less than 2 μM .