METHANOGENS IN ZEBU-CROSS CATTLE, IDENTIFIED PRESUMPTIVELY WITH IMMUNOFLUORESCENCE MICROSCOPY

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Methanogens that have been isolated from the rumen and from faeces of ruminants belong to the genera *Methanobrevibacter*, *Methanomicrobium* and *Methanosarcina* (Miller *et al.* 1986a). In this study immunofluorescence microscopy was used to identify presumptively methanogens in rumen contents that were available (preserved in formalin), from a study of the effects of monensin, avoparcin and formaldehyde-treated casein supplements on production in Zebu-cross heifers with *ad libitum* access to sorghum and supplementary molasses and minerals (Schlink *et al.* 1989).

Serum antibodies raised in rabbits to *Methanobrevibacter ruminantium* strain M1, *Methanobrevibacter smithii* strains PS, B181 and ALI-A, *Methanosarcina barkeri* strain MS, *Methanobrevibacter arboriphilus* strains DC, DH1 and AZ, *Methanobacterium formicicum* strain MF, and *Methanobrevibacter* species (strain Z6, Miller *et al.* 1986b) (but not strains of *Methanomicrobium*) were used as 'probes' in immunofluorescence microscopy (see Baker *et al.* 1998).

This is the first report of the likely composition of the rumen methanogens population in cattle using such a wide array of antibody 'probes'. Cells that resembled morphologically species and strains of *Methanobrevibacter* and to which antibodies bound strongly were the most commonly observed methanogens in these rumen samples (Table). *Mbr. smithii* has not been reported as an isolate from the rumen, but the observations here of cells that resembled *Mbr. smithii* morphologically and to which antibodies to strains of *Mbr. smithii* bound strongly are consistent with previous observations of rumen contents of cattle (with a smaller array of antidodies probes') and a sheep (Baker *et al.* 1998). Methanogens that were identified presumptively as *Mb. formicicum* and as one strain of *Mbr. arboriphilus* were found in only one group of cattle. Work is underway in our laboratory to use immunofluorescence microscopy and molecular biological techniques to presumptively identify methanogens isolated from the rumen of sheep and cattle using conventional culture techniques.

Diet	Methanogens species (presumptive identification)
Sorghum and molasses	Methanobrevibacter smithii strains PS
	Methanobrevibacter ruminantium strain M1
	Methanobrevibacter species strain Z6
Sorghum and molasses, plus 150mg avoparcin/head/day	Methanobrevibacter smithii strains PS, B181, ALI-A
	Methanobrevibacter ruminantium strain M1
	Methanobrevibacter species strain Z6
	Methanosarcina barkeri strain MS
Sorghum and molasses, plus 150mg monensin/head/day	Methanobrevibacter smithii strains PS, B181, ALI-A
	Methanobrevibacter ruminantium strain M1
	Methanobrevibacter species strain Z6
	Methanosarcina barkeri strain MS
Sorghum and molasses, plus 250g formaldehyde-treated casein/head/day	Methanobrevibacter smithii strains PS, B181, ALI-A
	Methanobrevibacter species strain Z6
	Methanobrevibacter ruminantium strain M1
	Methanosarcina barkeri strain MS
	Methanobrevibacter arboriphilus strain AZ
	Methanobrevibacter formicicum strain MF

Table 1. Species of methanogens identified presumptively in rumen contents from Zebu-cr	oss heifers	
from Townsville, Qld (pooled samples from 5 heifers per treatment group)		

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MILLER, T.L., WOLIN, M.J., HONGZUE, Z. and BRYANT, M.P. (1986b). *Appl. Environ. Microbiol.* **51**, 201-2. SCHLINK, A.C., GORDON, G.L.R., PHILLIPS<M.W. and SUTHERLAND, S. (1989) pp. 355-7 *In* 'The Roles of Protozoa and Fungi in Ruminant Digestion'. (Eds J.V. Nolan, R.A. Leng and D.I. Demeyer (Penambul Books: Armidale, NSW).