Ann Zootech (1995) 44, Suppl, 147

Rumen bacteria capable of growth on peptides and amino acids as sole source of energy : numbers and their role in ammonia production

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Excessive ammonia production by rumen micro-organisms is a major cause of inefficient N retention by ruminants. The micro-organisms which break down peptides and amino acids to ammonia were identified originally as typical major species of the rumen ecosystem (Bladen *et al,* 1961, Appl Microbiol, 9, 175-180), but more recently a smaller, more active population that used Trypticase (pancreatic casein hydrolysate containing peptides and some free amino acids) as their sole source of energy was identified (Chen and Russell, 1989, Appl Environ Microbiol, 55, 1052-1057). The aims of the present experiments were to enumerate the latter group of bacteria in the sheep rumen and to assess their role in ammonia production.

Four rumen-fistulated sheep received 500 g of hay, barley, molasses, fishmeal and vitamins-minerals (500, 299.5, 100, 91 and 9.5 g/kg DM) twice daily. Samples of rumen fluid were removed 2-3 h after feeding, strained, and diluted serially in the basal medium described by Chen and Russell (1990, Appl Environ Microbiol, 56, 2186-2192). These dilutions were used to inoculate Hungate tubes containing four different media : A, complete medium (Hobson, 1969, Meth Microbiol, 3B, 133-149) ; B, basal + 1.5 % Trypticase ; C, medium B + 5 µM monensin ; D, basal + 1.5 % casein acid hydrolysate. Growth was determined turbidimetrically after 7d incubation at 39°C and MPN was calculated from tubes reaching an OD>0.1. Ammonia production was measured in rumen fluid incubated *in vitro* with or without added 20 mg/ml Trypticase or amino acids with or without 5 µM monensin.

Bacteria growing on Trypticase comprised 1.5 % of the total viable count, but only 0.7 % grew with an OD>0.2. Most (93 %) of both categories were sensitive to monensin. Bacteria capable of growth on amino acids alone were 100-fold fewer in number. Less than half of total ammonia production, whether from endogenous protein, added Trypticase or amino acids, was inhibited by monensin. Hence numbers of monensin-sensitive bacteria capable of growth on Trypticase were low, but they had a significant, although minor, role in ammonia production by the mixed population.

	No addition		Monensin	
	Mean	SD	Mean	SD
Bacterial counts (x 10^7/ml)				
Total	188	130	ND	
Trypticase	2.8	1.9	0.2	0.1
Trypticase (OD>0.2)	1.4	1.5	0.1	0.2
Amino acids	0.03	0.01	ND	
Ammonia production (µmol/ml/h)				
No addition	0.85	0.72	0.58	0.26
Amino acids	3.73	1.09	1.92	0.70
Trypticase	4.35	1.00	2.47	0.63