EFFECT OF BASAL DIET AND FEED ADDITIVES ON THE SUSCEPTIBILITY OF SHEEP RUMEN FLUID TO LACTIC ACID PRODUCTION

J. B. Rowe, S. G. Winslow and P. J. Murray

Department of Agriculture, Baron-Hay Court, South Perth WA 6151, Australia

Introduction

Lactic acid production during rumen fermentation of carbohydrate can be a serious problem, under Australian conditions, in preparing animals for feeding pelleted diets during live export to the Middle East and during supplementary feeding programmes where cereal grain is trailed onto the ground.

There is a significant variation between animals in the amount of starch or soluble carbohydrate required to induce uncontrolled lactic acid production in the rumen and this makes the condition difficult to study in a quantitative way. At levels which give a high incidence of the condition, mortality among the more susceptible animals is a problem, while at levels which do not result in mortality, only 40 to 60% of animals develop high levels of lactic acid(Aitchison et al., 1987). In addition to between animal variation it is also likely that the basal diet affects the probability of developing acidosis. An alternative to dosing animals with starch or glucose is to remove samples of rumen fluid for *in vitro* incubation. This has the advantage that sufficient carbohydrate can be added for a predictable response without compromising the welfare of the animals.

Various antibiotic feed additives, active against Gram positive bacteria have been found to reduce lactic acid production during fermentation of starch or sugar in the rumen (Muir et al., 1980; Nagaraja et al., 1981; Aitchison et al., 1987). It is likely that these antibiotics act through the control of the major lactate-producing organisms, *Streptoccus bovis* and *Lactobacillus*. Feeding bentonite clays has also reduced stock losses during introduction of high starch grains (Dunn et al., 1979). The experiments reported here were designed to examine the effects of diet, bentonite clays and virginiamycin on acidosis, using an *in vitro* fermentation technique.

Materials and Methods

1. Evaluation of the in vitro technique

Variation between sheep and time of sampling. Twelve sheep (approx. 33 kg) were individually housed and fed a ration of chaffed cereal hay (900 g/d) for 17 weeks prior to the commencement of the experiment. Samples of rumen fluid were taken from each sheep using a stomach tube on days 1, 3 and 5 of the experiment and were incubated as described below. On days 1 and 3 the samples were taken prior to feeding and on day 5, three hours after feeding.

Effect of virginiamycin on lactic acid production. A sample of rumen fluid (500 ml) was obtained, following slaughter, from a sheep which had been grazing dry summer pasture and was strained through a nylon gauze (apertures approx. 200 μm). Incubations were carried out in triplicate with the following levels of virginiamycin: 0; 0.5; 1.0; 2.0 and 4.0 μg/ml.

2. Effect of diet and the inclusion of bentonite clay.

Thirty nine individually housed sheep were fed a diet of chaffed cereal hay (900 g/d), and an equal number received a pelleted diet (1400 g/d) containing (g/kg), lucerne chaff (590), lupin grain (250), barley grain (150) and a mineral/vitamin supplement (10). These diets were fed on their own (n = 12 animals) or with one of three types of bentonite clay (25 g/kg) (n=9/bentonite clay treatment) for a period of 9 weeks prior to samples of rumen fluid being taken, approximately 3 hours after feeding, for incubation with glucose.

In vitro incubation technique

Strained rumen fluid (16 ml) was added to a 25 ml McCartney bottle with 4 ml of glucose solution (60 mg/ml). The container was then flushed with CO_2, sealed and a 25 gauge needle inserted through the lid to allow gas to escape. Samples

AJAS 1989 Vol. 2 (No. 3) 346-348

were incubated at 38°C for 24 hours after which pH was measured and sample were taken for analysis of L-lactic acid concentration using a Cobas Mira Auto Analyser (Roche) and L-lactate kit (Behring).

Results

1. Evaluation of the in vitro technique

There were significant ($p < 0.05$) differences in the concentration of lactic acid following incubation of rumen fluid from different sheep. The observed range was between 33.2-70.3 mmol/L. There were no significant differences ($p > 0.05$) associated with sampling on different days (mean + SE: 39.4 + 2.81; 39.2 ± 5.04 and 51.8 ± 3.84 mmol L-lactate/L for days 1, 3 and 5 respectively) but there was a higher concentration of lactic acid produced in samples taken 3 hours post feeding (51.8 ± 3.84, day 5) compared to samples taken prior to feeding (39.3 ± 2.76).

Lactate concentration were reduced ($p < 0.001$) by virginiamycin at all levels studied in this experiment. Concentrations of L-lactate were (mean ± SE): 25.7 ± 2.2, 10.0 ± 2.4, 7.3 ± 2.4, 11.2 ± 2.4, 11.2 ± 2.4 and pH values were: 5.2 ± 0.58, 5.4 ± 0.58, 5.4 + 0.57, 5.3 + 0.55, 5.3 ± 0.58 for virginiamycin at 0, 0.5, 1, 2, and 4 μg/ml, respectively.

2. Basal diet and the effect of bentonite clay.

The results of the incubation of rumen fluid from sheep on two basal diets and containing no additive or one of three samples of clay is summarized in table 1.

There was a significant ($p < 0.01$) difference between diets. The rumen fluid from animals fed the pelleted diet had very much lower levels of lactic acid than those on the basal chaff diet. In animals on the chaff diet, clay A appeared to reduce the production of lactic acid ($p < 0.05$). The other two clays had no effect on lactic acid production *in vitro*.

TABLE 1. EFFECT OF BASAL DIET WITH OR WITHOUT BENTONITE CLAYS ON THE PRODUCTION OF L-LACTIC ACID (mmol/L)

	Chaff diet				Pelleted diet			
	Control	Clay A	Clay B	Clay C	Control	Clay A	Clay B	Clay C
n...	12	9	9	9	12	9	9	9
L-lactate	53.7	34.2	49.1	44.8	3.6	20.4	10.8	5.5
SE	3.84	6.91	3.36	3.63	0.42	7.43	3.69	1.06

Discussion

The variation of in vitro lactic acid production from different animals was expected from the *in vivo* studies reported by Aitchison et al. (1987). However it is clear that in samples taken at the same time with respect to feeding, similar results are achieved on different days. It therefore appears that this technique could be used to assess the susceptibility of an individual or a group of animals to carbohydrate overload and lactic acidosis. The increase in lactic acid production from samples taken post feeding indicates that the practice of starving animals prior to studies on lactic acidosis may not be advisable.

It is clear that the antibiotic virginiamycin controlled lactic acid production even at concentration of 0.5 μg/ml. That the pH fell, irrespective of the extent of lactic acid production, suggests that total volatile fatty acid (VFA) production was not affected by the antibiotic.

There was a clear effect of the basal diet on the rumen fluid's ability to produce lactic acid *in vitro*. Lactic acid only accumulates if the rate of production exceeds the rate of utilization or conversion to VFA. It can therefore result either from a proliferation of lactate producers or a failure of lactate utilisers to proliferate rapidly enough to utilise the increased quantity of the acid. On the pelleted diet more readily fermentable carbohydrate was provided than on the chaff diet and it could therefore be argued that there would be a greater population of lactate utilisers in the rumen fluid of these animals than on the chaff diets. It is suggested that the effect of the basal diet on lactate accumulation is through the presence or absence of lactate-utilizers, whereas the effect of feed additives is through the control of the lactate-producers.

The bentonite clays (estimated concentration, approx. 2.5 mg/ml) did not have an appreciable effect on lactic acid production compared to the antibiotic virginiamycin (0.5 μg/ml). The result with respect to the efficacy of bentonite relative to the antibiotic virginiamycin, is similar to that reported by Aitchison et al. (1987) for bicarbonate relative to avoparcin and suggests that the antibiotic feed additives offer a far greater potential than bentonite or buffers in controlling acidosis during the introduction of ruminants to high-grain diets.

(**Key Words**: Acidosis, Bentonite, Virginiamycin)

Literature Cited

Aitchison, E.M., C.L. McDonald, P. Casson and J.B. Rowe. 1987. Feed antibiotics and buffers to control rumen lactic acid. Proc. Nutr. Soc. 46:41A.

Dunn, B.H., R.J. Emerick and L.B. Embry. 1979. Sodium bentonite and sodium bicarbonate in high concentrate diets for lambs and steers. J. Anim. Sci. 48:764-769.

Muir, L.A., E.L. Rickes, P.F. Duquette and G.E. Smith. 1980. Control of wheat induced acidosis by thiopeptin and related antibiotics. J. Anim. Sci. 50:547-553.

Nagaraja, T.G., T.B. Avery, E.E. Bartley, S.J. Galitzer and A.D. Dayton. 1981. Prevention of lactic acidosis in cattle by lasalocid or monensin. J. Anim. Sci. 53:206-215.