A STUDY ON THE EFFICACY OF ANTI-PROTOZOAL MOLASSES BLOCKS IN SHEEP

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by

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PREFACE

The studies presented in this thesis were completed by the author while a post graduate student in the Department of Animal Science, the Faculty of Science, the University of New England, Armidale, N.S.W., Australia Assistance given by other persons is indicated in the text or in the list of acknowledgments. All references cited are included in a bibliography. The work is otherwise original.

* * *

I certify that the thesis has not already been submitted for any degree and is not being currently submitted for any other degree.

I certify that any help received in preparing this thesis, and all sources used, have been acknowledged in this thesis.

February, 1997 *Nani Sufiani Suhanda*

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SUMMARY

The absence of ciliate protozoa from the rumen is known to reduce proteolysis of dietary protein in the rumen and to increase the supply of microbial protein for digestion by the host, thereby increasing the quantity of amino acids available for absorption. Therefore defaunation can be expected to improve production in ruminants fed low-protein diets. However, no practical method of controlling protozoa in the rumen of grazing animals has been developed. The current studies were initiated to examine the efficacy of molasses blocks containing an anti-protozoal agent in reducing or completely removing protozoa from the rumen of sheep. The detergents Teric GN9 and Alkanate $3SL_3$ were used as anti-protozoal agents because it had been demonstrated that these detergents had successfully defaunated the rumen when administered as an oral drench. Preliminary studies indicated that blocks containing molasses up to (67 %), wheat bran (25%), lime (8%) and up to 10% detergent set sufficiently hard to prevent sheep from over-consuming block.

A. *In vivo* studies

1. A basal diet (90% oaten chaff, 10 % barley grain and 10 g urea/d) was given *ad libitum* to twelve Merino wethers as experimental animals. Molasses blocks containing 5% and 10% of Alkanate 3 SL_3 and Teric GN9 were tested, and the voluntary intakes of the basal diet and the molasses blocks, the diurnal pattern of block intake, and the population of protozoa were monitored.

i) The mean daily intake of blocks containing Teric was 28 ± 4 g/d, whereas the mean daily intake for blocks containing Alkanate was 161 ± 26 g/d. This result suggests that Teric was less preferred than Alkanate. In the first 3 weeks of treatment, blocks containing 5% Alkanate were consumed at a greater rate than those containing 10 % Alkanate (218 ± 31 vs. 121 ± 15 g/d; $P<0.05$) and thereafter were similar (148 ± 13 g/d). The diurnal measurements of block intake showed that the highest rate of block intake (22 g/h) occurred in the morning between 9.30 am and 12.30 am, when animals consumed the major portion of their daily feed. At this time the detergent would be expected to be least effective due to dilution effects of saliva and possibly of adsorption of detergent onto fibre.

iii) The key finding was that molasses blocks containing anti-protozoal detergent were not effective in eliminating protozoa from the rumen of sheep. The mean protozoal populations for sheep receiving Teric GN9 and Alkanate 3 SL3 were 2.59 ± 0.33 and 1.70 ± 0.27 x 10^5 cells/ml respectively compared with 2.1 ± 0.3 x 10^5 cells/ml in sheep. There were no significant differences between types or level of detergent with respect to their effects on the rumen protozoal population. The failure of Teric GN9 to control rumen protozoal numbers was almost certainly to due to the low daily intake (2.8 ± 0.4 g/d) of this detergent. The reason for the failure of Alkanate 3 SL3 to control rumen protozoa was less obvious as the average daily intake of this detergent was 11.37 ± 0.89 g/d

which was expected to be sufficient to defaunate the rumen. Further studies were undertaken to investigate possible reasons for the failure of Alkanate 3 SL3 to defaunate the rumen.

2. The influence of feed deprivation on the efficacy of anti-protozoal molasses blocks in removing protozoa from the rumen was investigated. Two levels of Alkanate 3 SL_3 (5% and 10%) were tested. Block intakes and protozoal population were monitored before and during feed restriction.

   i) Molasses blocks containing both 5% and 10% Alkanate 3 SL_3 successfully defaunated sheep when combined with a 4-day period without access to the basal diet. Protozoal population declined from $2.58 \pm 0.25 \times 10^5$ cells/ml on full feed to an undetectable level on day 4 of feed restriction.

   ii) The level of Alkanate in the molasses block during feed deprivation significantly affected block consumption (169 ± 71. 11 ± 6 and 43 ± 24 g/d for 5%, 10% and 0 % Alkanate respectively).

B. *In vitro* studies

Two possible causes of the failure of molasses blocks containing Alkanate 3 SL3 to remove protozoa from the rumen of sheep were investigated through a series of *in vitro* experiments. The level of fibre in the rumen and the age of the Alkanate used were identified as the most likely factors involved. Because of the replication and control possible, an *in vitro* approach was adopted. To maintain the consistency of the incubation medium, an artificial rumen fluid (RF^+ medium) was used in preference to fresh rumen fluid, which may change over time.

3. Under *in vitro* conditions the anti-protozoal activities (expressed as a % reduction in protozoal numbers) of old Alkanate 3 SL_3 and the fresh Alkanate 3 were 73.44 ± 3.10 % vs. 69.64 ± 2.52 % respectively This result indicated that even though the Alkanate $3SL_3$ used in Experiment 1 and 2 was 6 years old it still retained its full anti-protozoal activity compared to fresh Alkanate. It was concluded that age of detergent was not a contributing factor in the failure of Alkanate to defaunate the rumen in Experiment 1.

4. Oaten chaff used in Experiment 1 was finely ground and added to the incubation medium at two levels: 0.2 and 0.4 g/ 10 ml. In the absence of fibre Alkanate reduced protozoal number by 84%. In the presence of 0.02 and 0.04 g/ml of fibre Alkanate reduced protozoal number 52 % and 50 % respectively; indicating that the presence of fibre resulted in a significant ($P< 0.001$) reduction in the anti-protozoal activity of the detergent. This result and the results obtained in Experiment 2, suggest that one of the main causes of the failure of anti-protozoal molasses blocks to defaunate sheep in Experiment 1 was a reduction of detergent effectiveness due to the adsorption of active molecules onto feed particles.

Results from these studies demonstrated that administration of anti-protozoal detergents via molasses block were unable to defaunate the rumen of sheep fed a roughage diet *ad libitum*. Therefore anti-protozoal molasses block containing detergent are unlikely to be successful in controlling protozoa in grazing ruminants.